JPRS L/8481

25 May 1979

TRANSLATIONS ON USSR SCIENCE AND TECHNOLOGY

BIOMEDICAL AND BEHAVIORAL SCIENCES

(FOUO 17/79)

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APPROVED FOR RELEASE: 2007/02/09: CIA-RDP82-00850R000100050052-8

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Moscow DOKLADY VSESOYUZNOY ORDENA LENINA AKADEMII SEL'SKOKHOZYAYSTVENNYKH NAUK IMENI V.I. LENINA in Russian No 12, 1978
Nos 1 and 3, 1979

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AGROTECHNOLOGY

UDC: 633.11"324":632.111

EFFECTS OF LOW AND HIGH TEMPERATURES IN THE SPRING ON DURATION OF VEGETATION PERIOD AND YIELD OF WINTER WHEAT

Moscow DOKLADY VSESOYUZNOY ORDENA LENINA AKADEMII SEL'SKOKHOZYAYSTVENNYKH NAUK IMENI V. I. LENINA in Russian No 12, 1978 pp 1-3

[Article by A. I. Korovin, doctor of biological sciences, and G. I. Kozlov (presented by K. Z. Budin, academician of the All-Union Academy of Agricultural Sciences imeni Lenin on 14 Mar 77), All-Union "Order of Lenin" and "Order of People's Friendship" Scientific Research Institute of Plant Growing imeni N. I. Vavilov, submitted 11 Apr 77]

[Text] We know that there is a fluctuation in yield of winter crops in areas with adequate humidity [1, 4-6] because of weather conditions in the autumn-winter, winter-spring and summer periods. It was established that, in the case of good wintering, the harvest is largely determined by the temperature conditions in the first 20-30 days of spring. With moderately low temperatures (8 to 12°C), a higher harvest is formed than at low (6-7°C) or higher (18-20°C) temperatures. However, it is not clear how winter plants react to high and low temperatures over short periods of time.

To determine this, experiments were conducted in 1975 and 1976 with Mironovskaya 808 variety of winter wheat at the Pushkin laboratories of the VIR [All-Union Scientific Research Institute of Plant Growing], in vegetation containers that could hold 5 kg absolutely dry sandy loam, fertilized with NPK at the rate of 0.1 g of each nutrient element per kg dry soil. Winter wheat was sown in late August. The plants were under identical conditions in the fall, they produced up to 2-3 shoots before the end of vegetation and wintered well in a special plot [2].

In the spring, when vegetation resumed, the plants were thinned out. We left 10 plants per container and added mineral fertilizers. In the middle of May, when the plants evened out, all of the containers were divided into three groups and placed in air-temperature controlled vegetation buildings. Specific temperatures were maintained in these buildings for 30 days. In the first group (control) mean daily temperature was 18°C (ranging from 15°C at night to 21°C in the daytime); in the second it was 10°C (4°C at night to 16°C in the daytime) and in the third, it was 26°C (20°C at night and 32°C in the daytime).

Table 1. Effect of low and high temperatures on start of developmental phases in winter wheat

Temp. °C after resumption of vegetation (°C)	Duration of exposure (days)	Duration of period (days): resumption of "shooting"	"shooting" heading	heading—milk-ripe stage	milk-ripe—yellow-ripe	resumption of veget.—heading	heading—maturation	resumpt. of vegetation—maturation
18	30	20	32	24	8	52	32	84
10	5	20	33	25	8	53	33	86
	10	20	34	26	9	54	35	89
	15	20	34	28	9	54	37	91
	20	20	37	30	11	57	41	98
	25	20	40	32	13	60	43	103
	30	20	45	34	13	65	47	112
26	5	20	31	22	8	51	30	81
	10	20	31	21	8	51	29	80
	15	20	30	20	7	50	27	77
	20	20	28	19	7	48	26	74
	25	20	26	18	6	46	24	70
	30	20	24	17	6	44	23	67

Table 2. Effect of low and high temperatures on winter wheat harvest and structure thereof

Temperature (°C) after resumption of vegetation	Duration of exposure	Harvest: total g/container	total %	grain g/container	grain %	straw g/container	straw %	grain/straw ratio	% grain in harvest	grain per spike	weight per 1000 grains (g)	Bushiness: total	productive	plant height (cm)
18	30	75,5	100	29,4	100	46,1	100	0,64	39	23	36	3,2	3,0	90
10	5	76,7	101	30,2	103	46,5	101	0,65	39	24	36	3,2	3,0	90
	10	78,8	102	30,5	104	46,3	100	0,65	40	24	37	3,4	3,0	88
	15	77,8	103	31,9	108	46,0	100	0,69	41	23	37	3,5	3,2	90
	20	80,8	107	33,4	114	47,4	103	0,70	41	25	38	3,7	3,2	90
	25	84,8	112	35,3	120	49,5	107	0,71	42	26	38	3,8	3,4	90
	30	87,3	116	37,1	126	50,2	109	0,74	42	26	38	4,4	3,5	90
26	5	67,9	90	23,6	80	44,3	96	0,53	35	21	35	3,3	3,1	83
	10	64,4	85	20,4	69	44,0	95	0,46	32	20	35	3,8	2,9	70
	15	60,4	80	19,2	65	41,2	89	0,47	32	19	32	4,0	2,8	70
	20	47,4	63	9,6	33	37,8	82	0,25	20	16	28	4,4	2,0	63
	25	45,0	60	5,7	19	39,3	85	0,14	13	12	28	4,5	2,3	59
	30	47,3	63	5,2	18	42,1	91	0,12	11	12	23	5,4	1,6	55

In all of the variants, soil humidity was kept at 50-60% of its total moisture capacity. Four containers from the 2d and 3d buildings were transferred to the first one 5 days after the start of the experiment. This was repeated after 10, 15, 20, 25 and 30 days. After 30 days, all of the containers

were placed in a mesh-enclosed building (for protection against birds), where they were kept until harvesting time under the same conditions.

Table 3. Daily productivity of winter wheat as related to low and high temperatures

Temp. (°C) after resumption of vegetation	Duration of exposure (days)	Days from resumed vegetation to maturation	Mean harvest increment per vegetation day			
			total		grain	
			g/contain.	%	g/contain.	%
18	30	84	0,89	100	0,35	100
10	5	86	0,89	100	0,35	100
	10	89	0,89	100	0,34	197
	15	91	0,86	96	0,35	100
	20	98	0,82	92	0,34	97
	25	103	0,82	92	0,34	97
	30	112	0,78	88	0,33	94
26	5	81	0,84	94	0,29	83
	10	80	0,80	90	0,25	83
	15	77	0,78	88	0,25	71
	20	74	0,64	72	0,13	37
	25	70	0,64	72	0,08	22
	30	67	0,71	80	0,08	22

Table 1 shows that there was distinct manifestation of the effects of low and high temperatures already within 5 days, and it progressed at each subsequent interval. In the case of 30-day exposure to low temperatures, maturation occurred 28 days later and with exposure to high temperatures, 17 days earlier than in the control. The difference between the extreme variants was 45 days.

As can be seen in Table 2, the temperature level in the spring had the least effect on yield of straw. There was a significant difference between variants with respect to overall harvest: it increased by 16% with low temperatures and decreased by 40% with high ones. The temperature levels had the strongest effect on grain harvest, which increased by 26% in the case of 30-day exposure to low temperatures and decreased by over 80% with exposure to high temperatures, as compared to the control.

The grain to straw ratio, percentage of grain in the overall harvest and plant height were also lowest in these variants.

The decrease or increase in grain harvest under the influence of low and high temperatures is attributable to changes in number of fruit-bearing stems, grain content per spike and grain size (Table 2). It is important to mention

that the increment in harvest, particularly grain, was higher on each day of vegetation at low temperatures than at high ones (Table 3).

All of these data were obtained from model experiments. They simulated the mean extreme variants of temperatures, which do not occur every year. It must be borne in mind that the experiments were conducted during the period of white nights. In the South, where there is a longer night, the absolute results will apparently be different, but their direction will be the same. Even brief exposure to higher (not high) or lower temperatures has an effect. It is imperative to study, by means of model experiments, the most important situations of interaction between plants and specific meteorological factors in order to ultimately learn all about the changing process of harvest formation as related to weather conditions.

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CSO: 1870

AGROTECHNOLOGY

UDC: 633.11:581.154

APPEARANCE OF SPHEROCOCCOIDS WITH INDUCED MUTAGENESIS

Moscow DOKLADY VSESOYUZNOY ORDENA LENINA AKADEMII SEL'SKOKHOZYAYSTVENNYKH NAUK IMENI V. I. LENINA in Russian No 12, 1978 pp 3-6

[Article by G. V. Ryskal' and I. D. Mustafayev, academician of the Azerbaydzhan Academy of Sciences, Institute of Genetics and Breeding, Azerbaydzhan Academy of Sciences, submitted 3 Feb 78]

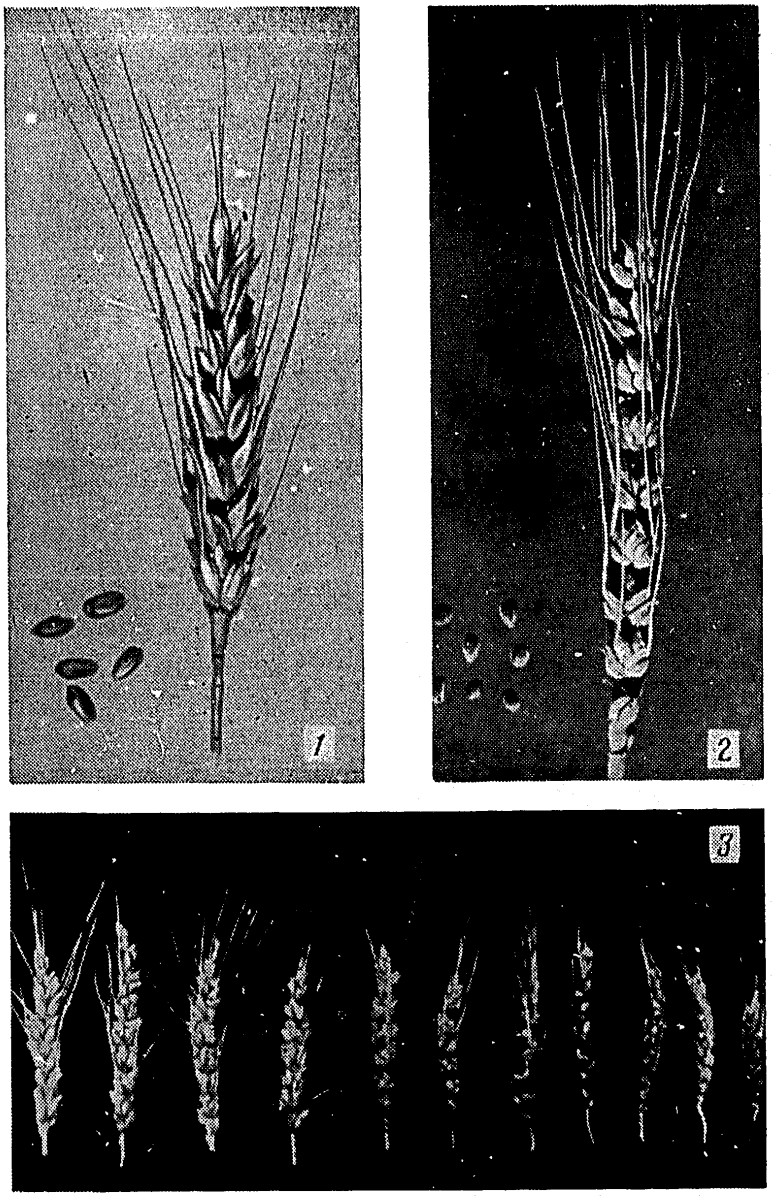
[Text] After using chemical mutagens on breat wheat [T. aestivum L.], along with development of economically valuable mutants, our objective was to obtain spherococcoid mutants and continue our study of inheritance of this character.

There are reports in the literature of occurrence of spherococcoid mutants in bread wheat after induced mutagenes; however, they presented segregation of some characters of this complex (firmness of spike, height of plants, spherical grain). I. S. Morozova et al. [2], in particular, who treated seeds of Belotserkovskaya 198 bread wheat seeds with N-nitrosoethylurea, obtained a large number of mutants differing from the original form in only one character, spherical grain. The genetic analysis made by the authors revealed that the spherical grain character is dominant and controlled by one gene.

Other authors [1, 3, 4] discovered spherococcoid mutants in hard wheat [T. durum] and assumed that the individual genes determining this spherococcoid character are either in genome A or genome B.

The Bulgarian researchers, Georgiev and Nicolov [5], who used EMS (ethylmethane sulfonate), obtained a mutant with the phenothpe of round-grain wheat in one-grained wheat [Triticum monococcum L.], which also confirms the hypothesis that there are individual genes for spherical grain in other genomes, in this instance genome A.

In the studies of N. N. Zoz [1], five mutants with round grain, other characters being normal, and two mutants with a dull-tipped, hard and broad leaf were obtained in various cultivars of winter bread wheat under the influence of N-nitrosomethylurea (NMM), along with mutants with the set of characters typical of spherococcum. These mutants appeared in M_1 and were notable for normal fertility; however their offspring was unviable due to change to a homozygous state of mutated genes.



Effect of polymer genes on appearance of spherococcoid mutants: 1) spike and grain of initial Gyurgyan 3 bread wheat; 2) spike and grain of spherococcoid mutant isolated in M_2(NMM, 0.02% concentration); 3) spikes with different degrees of manifestation of spherococcoid character in M_3.

Normal phenotype (bread wheat)--$S_1S_1S_2S_2$

Mutange NMM (concentration 0.02%) →

sharp changes in day and night temperatures, increased solar insolation, etc.

→M_2 mutation of S_1, S_2 loci occurs -- $S_1s_1S_2s_2$ intermediate type with characters of bread wheat and spherococcum

M_3 segregation of the polymerism type:

1.	$S_1S_1S_2S_2$	0%	spherococcoid character	normal phenotype of bread wheat
2.	$S_1S_2S_1s_2$	25%	" "	phenotype with noticeable signs of spherococcoid features
3.	$S_1S_2s_1S_2$	25%	" "	same
4.	$S_1S_2s_1s_2$	50%	" "	phenotype with intensified spherococcoid character
5.	$S_1s_2S_1s_2$	50%	" "	same
6.	$S_1s_2s_1s_2$	75%	" "	intensification of spherococcoid character
etc.				
...	$s_1s_1s_2s_2$	100%	" "	pure spherococcum

We conducted experiments in a mountain region (Kedabekskiy Rayon, Azerbadzhan SSR) characterized by a cold climate. The summer is short, moderately warm and the winter is dry. The mean annual air temperature over a period of many years constituted +7.4°C. Annual precipitations constitute 530 mm. Mean annual relative air humidity is 72%. The soil is of the forest-ore [or mountainous], humus carbonate type.

Dry seeds of Gyurgyana 3 (v. erythrospermum) bread wheat were treated with the chemical mutagen NMM in a concentration of 0.02% (exposure for 20 h). Treated seeds were sown in three different regions (climate, soil, vertical zonality, 80, 420 and 1750 m above sea level).

In M_2 we found altered forms with some signs of spherical-seed wheat (short straw, inflated spikelet glume, round grain that is almost spherical). These spherococcoid plants are referable to the second type of phenotype [3]. The incidence of spherococcoids in M_2 under the influence of NMM on bread wheat constituted 2.0%, whereas that of speltoids was 9.5% and multiflorous 8.2% with the same treatment variant. We did not observe spontaneous spherococcoids.

A 1:13 segregation was observed among the offspring of spherococcoid plants (M_3), where there were 13 spherococcoid plants with varying degrees of roundness of grain and inflation of spikelet glume per plant of the original bread wheat type, representing the gradual transition from bread wheat to spherical-grain wheat (Figure). All this warranted the belief that the action of two pairs of polymeric genes was involved.

Inheritance of red-colored spike and grain in some wheat hybrids is one example of interaction of polymeric genes. The Swedish scientist Nilsson-Ehle [6] expounded a hypothesis of identical factors, i.e., dependence of one character on several genes affecting the character in the same direction and with the same force. Let us assume that the normal phenotype (original form) is determined by two pairs of identical genes $S_1S_1S_2S_2$, each of which induces, in the recessive state, a certain degree of development of the spherococcoid character.

Let us stipulate that the action of each gene constitutes 25%, and let us describe schematically inheritance of spherococcoid sign with mutation of S_1 and S_2 loci.

Thus, for complete expression of the spherococcoid character there must be two pairs of recessive genes present (the dominants will yield the pure original form, i.e., bread wheat, and the recessive will yield a pure spherococcum). Different combinations of identical genes lead to enhancement of one character and attenuation of the other. The degree of development of the character depends on the number of identical genes in the zygote, be they in homozygous or heterozygous state. In our experiments, we failed to demonstrate segregation of a pure spherococcum, since the S_2 block [unit] is in a semidominant state.

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10,657
CSO: 1870

AGROTECHNOLOGY

UDC: 636.086.15:631.8

DEPENDENCE OF GREEN MASS QUALITY OF CORN ON AREA OF NUTRITION AND FERTILIZER BACKGROUND IN SIBERIA

Moscow DOKLADY VSESOYUZNOY ORDENA LENINA AKADEMII SEL'SKOKHOZYAYSTVENNYKH NAUK IMENI V. I. LENINA in Russian No 12, 1978 pp 6-7

[Article by A. N. Vasil'yev (presented by I. I. Sinyagin [deceased], academician of the All-Union Academy of Agricultural Sciences imeni Lenin, on 4 Jan 78), Siberian Scientific Research Institute of Agriculture, submitted 17 Jan 78]

[Text] Concurrent change in level of mineral nutrition and density of plant stands constitutes a complex factor that forces plants to utilize differently solar radiation, soil and atmospheric moisutre, soil nutrients and fertilizers, etc. All this ultimately affects not only the quantitative, but qualitative aspect of the production process.

Experiments dealing with the joint effect of nutrition area and level of mineral fertilizers ($N_{90}P_{90}K_{60}$) on qualitative parameters of green mass of Sterling variety corn were conducted in 1970-1972 in the northern forest-steppe region near the Ob' River, at a base of the Siberian Scientific Research Institute of Plant Growing and Breeding.

Corn was raised on a nutrition area of 20×20 cm (250,000 plants per ha [hectare]), 40×40 cm (52,500), 60×60 cm (27,800), 80×80 cm (15,600), 100×100 cm (10,000) and 120×120 cm (7000). The variants were repeated four times. The plot size changed from 52 to 280 m^2, with increase in nutrition area. Annual grain crops were the precursors.

The experiments were conducted on the typical soil of the area, moderately loamy chernozem, which has a mildly acid reaction, with 6.3% humus in the 0-20 cm layer (according to Tyurin), 0.308% total nitrogen, 22 mg mobile phosphate and 18.5 mg potassium per 100 g dry soil. The soil was treated in the usual way for the region. The corn was planted with a hand sower, at the rate of 2-3 grains per pit, and after sprouting, one plant was left in each. We determined nitrogen, phosphorus and potassium content of the plants by means of rapid ashing of a suspension in concentrated sulfuric acid; total nitrogen was assayed according to Kjeldahl, phosphoric

acid was determined colorimetrically, potassium with flame photometry wet ash by dry ashing and wet cellulose by the method of Henneberg and Stoman.

Feed unit and digestible protein content (kg/ha) of green corn mass as related to nutrition area and fertilizers (in different years)

Experimental variant (cm)	No fertilizers				$N_{90}P_{90}K_{60}$			
	1970	1971	1972	mean	1970	1971	1972	mean
				Feed units				
20×20	4325	5183	6174	5227	8357	7484	7794	6878
40×40	2995	3657	4295	3699	4227	5808	5411	5149
60×60	2763	3502	2812	2689	2693	3614	2251	2853
80×80	1796	1884	1731	1804	1868	2403	2024	2098
100×100	1009	1164	1275	1149	1399	1649	1852	1633
120×120	748	1090	974	936	765	1109	1120	998
				Digestible protein				
20×20	447	443	410	433	566	657	504	576
40×40	306	274	317	299	459	440	569	489
60×60	277	382	263	291	353	367	250	323
80×80	223	227	240	230	250	240	230	240
100×100	131	159	114	135	182	152	167	167
120×120	98	124	115	112	130	109	126	122
				Digestible protein per feed unit				
20×20	103	86	66	85	106	88	68	87
40×40	103	71	84	86	109	76	105	97
60×60	101	141	94	112	131	101	111	114
80×80	124	120	139	128	134	100	114	116
100×100	129	136	84	116	130	92	90	104
120×120	131	114	118	121	169	98	112	126

Estimates of yield of feed units and digestible protein per hectare corn revealed that the levels fluctuated markedly, depending on the nutrition area and fertilizer background (Table). The highest number of feed units (5227 kg/ha) and largest amount of digestible protein (433 kg/ha) were obtained in the experiments without addition of fertilizers in the variant with a nutrition area of 20×20 cm (250,000 plants/ha). Addition of complete mineral fertilizer in this variant, in a dosage of $N_{90}P_{90}K_{60}$, increased the feed unit yield to 6878 kg/ha and digestible protein to 576 kg/ha. But availability of digestible protein per feed unit was the lowest in this variant: 85 g without fertilizers and 87 g with them. This is apparently attributable to two causes; in the first place, reduced light in the dense stands and low nitrate content of soil [1, 2] and, in the second place, mild effect of fertilizers since, in this case, the N_{90} dosage is not sufficient to provide for nitrogen nutrients for the plants. For this reason, the dosage of nitrogen must be increased in the adopted $N_{90}P_{90}K_{60}$ combination of fertilizers.

Thus, in spite of some decrease in quality of green mass yield from corn in the case of higher planting density, we can recommend expressly such density, since the overall yield of feed units and digestible protein is

high per unit area. The protein deficiency should be corrected by means of appropriate nitrogen supplements.

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10,657
CSO: 1870

AGROTECHNOLOGY

UDC: 633.26/29:636.085.2

ENERGY AND PROTEIN CONTENT OF GRASS AND CONFORMITY THEREOF WITH THE REQUIREMENTS OF A HIGHLY PRODUCTIVE DAIRY HERD

Moscow DOKLADY VSESOYUZNOY ORDENA LENINA AKADEMII SEL'SKOKHOZYAYSTVENNYKH NAUK IMENI V. I. LENINA in Russian No 12, 1978 pp 14-16

[Article b y Kh. M. Older, candidate of agricultural sciences (presented by N. G. Andreyev, academician of the All-Union Academy of Agricultural Sciences imeni Lenin on 23 Dec 77), Estonian Scientific Research Institute of Agriculture and Reclamation, submitted 16 Dec 77]

[Text] The energy and protein content of feed is very important. It is considered that of the overall value of feed about 50% is referable to energy, 20-30% to protein and 20-30% to other components.

For a long time, the chief gage of nutrient value in feeding practice was the feed unit. Since the 1960's, there has been a change in several countries (Great Britain, Sweden) to a new system of determining the nutrient value of feed, based on calories or joules of metabolic energy.

The feed unit of oats is still used in the Soviet Union. At the present time, a new system is being developed [2, 4, 5], which is based on joules of metabolic energy (1 cal = 4.1868 J).

We studied the energy and protein content of gramineous grass, as well as in leguminous-gramineous grass mixtures. For this purpose, the gramineous grass was crossed for the first time at the tillering, booting, heading and flowering stages. In determining the time for mowing leguminous-gramineous mixtures, we took into consideration development of clover, and the first mowing was performed at the stages of stem formation, budding, start of flowering and complete flowering. The next mowing of all grass stands was performed 20-25, 35-40, 50-55 and 60-65 days after the preceding one.

The coefficients used to calculate the types of energy of digestible protein were taken from the tables used in GDR and in the department of livestock feeding of the Estonian Agricultural Academy [1, 6].

The experimental results indicate that the concentration of metabolic energy of grass depended mainly on the phase of development and type of grass.

Table 1. Amount of digestible protein per unit metabolic energy (g/MJ) as related to stage of grass development and level of fertilization (mean data for 1974-1975)

Stage of grass development	Common foxtail	Cocksfoot	Meadow fescue	Reed canary grass	Awnless brome	Timothy grass	Red clover with Timothy grass	Leguminous-gramineous pasturage
	$N_{84}P_{66}K_{66}$ first mowing						$N_0P_{66}K_{66}$	
Tillering (stem-form) (I)	14,1	13,9	12,2	17,2	13,9	9,8	13,6	12,7
Booting (budding) (II)	14,8	10,0	11,2	13,1	11,7	9,1	9,3	8,8
Heading (start of flowering (III)	9,5	6,7	8,1	6,9	9,3	5,7	6,4	6,7
Flowering (IV)	7,9	5,5	6,0	6,4	5,3	4,1	5,7	5,7
	$N_{200}P_{166}K_{330}$						$N_0P_{66}K_{66}$	
I	14,1	15,5	15,3	19,8	11,7	11,2	11,0	10,3
II	15,0	13,1	11,2	16,5	15,0	11,2	9,3	8,8
III	11,5	8,3	9,8	8,8	10,0	8,8	6,0	6,7
IV	11,7	5,5	6,7	8,1	6,0	5,3	6,4	5,0
	Means for 1-4 mowings							
	$N_{133}P_{66}K_{66}$						$N_0P_{66}K_{66}$	
I	10,3	10,3	11,2	12,2	8,7	10,7	11,5	10,0
II	10,7	9,8	9,3	11,2	8,8	10,3	9,3	9,5
III	9,3	7,9	7,9	8,3	8,8	7,2	8,3	8,1
IV	8,3	7,2	6,4	6,4	6,0	4,8	6,0	6,9
	$N_{200}P_{133}K_{330}$						$N_{70}P_{66}K_{66}$	
I	11,7	11,9	11,7	14,6	9,8	12,4	10,3	10,3
II	12,2	12,9	10,5	14,6	11,2	11,2	10,0	9,8
III	11,7	10,3	11,0	9,8	10,3	9,3	7,2	7,9
IV	11,7	7,9	7,9	9,3	6,9	6,0	6,9	6,2

The metabolic energy content of grass fluctuated over a relatively wide range; it was highest at the tillering stage (10.13-12.56 MJ/kg) and lowest at the flowering stage (6.91-9.84 MJ/kg). The top range of concentration of metabolic energy determined by the stage of grass development coincided approximately with the corresponding index for barley and oat meal and the bottom range, with the corresponding index of feed straw.

The species and mixtures studied can be listed in the following order of decrease in concentration of metabolic energy: meadow fescue, Timothy grass,

late-maturing red clover with Timothy, leguminous-gramineous pasturage mixed grass, common foxtail, cocksfoot, awnless brome and reed canary grass.

The protein content of grass depended significantly on the stage of plant development: the later the mowing, the lower the content thereof in dry substance. A radical decrease in protein content was noted at the late stages of development, particularly in reed canary grass, awnless brome, Timothy grass and leguminous-gramineous grass mixtures. When the first mowing was performed at the tillering stage, there was over 20% crude protein in dry grass in most cases, 14-20% at the booting stage and 9-14% at the heading stage. At the flowering stage, crude protein content of dry substance exceeded 14% only in awnless brome, and did not reach 12% in the other species.

In most cases, increased doses of fertilizers ($N_{300}P_{\omega 80}K_{270}$) caused a 2-4% increase in protein content of gramineous grass. Use of moderate doses of fertilizers ($N_{70}P_{60}K_{90}$) failed to induce changes in protein content of leguminous-gramineous grass mixtures.

Feed prepared from field grass with a large amount of clover is generally considered to be rich in protein. The results of the experiments indicate that the protein content was relatively high (13-18% in most cases) in dry substance of both field and pasturage grass only in the period preceding flowering. A sharp drop in protein content was observed with aging of the plants. Thus, dry substance contained 9-12% crude protein at the start of flowering and 8-11% at the stage of full bloom.

Balanced energy and protein in feed rations is very important to better assimilation of feed. Heretofore, this ratio was expressed as the amount of digestible protein (in grams) per feed unit.

In the last few years, new factorial and summary standards were developed in the department of livestock feeding of the Estonian Agricultural Academy with respect to energy and protein requirements as related to the age of the animals, level of productivity of dairy cows and type of feeding [2, 3].

Using the summary zootechnical norms as the basis, it can be calculated that at least 8.7 g digestible protein is required per 1 MJ metabolic energy for a balanced diet.

Table 1 lists the amounts of digestible protein per unit energy; the dash lines indicate the above-mentioned proportion of protein and energy. The figures above the dash lines refer to an abundance of protein in the feed and those under the dash lines, to a shortage thereof.

The results of analysis of samples taken from pure grass plantings revealed that one must cut down cocksfoot, meadow fescue, reed canary grass and Timothy grass no later than the booting stage in the case of using small doses of nitrogen fertilizer (N_{34}). The level of digestible protein per unit energy was adequate as well in the tillering phase only in the dry

substance of common foxtail and awnless brome. At the flowering stage, none of the grass species studied presented a balanced protein and energy content.

With the use of moderate doses of nitrogen (N_{100}), there was an increase in protein content of dry substance at all stages of grass development. The higher nitrogen content of the fertilizer made it possible to postpone the first mowing by one phase of development (2 weeks). This made it possible to mow the gramineous grass at the heading stage as well, but not at the flowering stage. In order to have a high protein and energy ratio, with moderate doses of fertilizers (total of N_{100} for the summer), the aftermath must be cut down at the age of 36-40 days, with the exception of awnless brome and common foxtail, in which this is present at 50-55 days. But the aftermath of the above-mentioned species is severely stricken by fungus diseases within such a vegetation period, so that it is desirable to mow them down at the age of 35-40 years.

Table 2. Amount of digestible protein as related to metabolic energy (g/MJ) against the background of $N_{100}P_{60}K_{90}$

Stage of grass development	Common foxtail	Cocks-foot	Meadow fescue	Reed canary grass	Awnless brome	Timothy grass
Tillering	14.6	13.4	13.9	13.4	16.8	11.9
Start of booting	13.6	13.1	14.8	14.6	15.3	12.9
End of booting	15.0	11.2	11.9	17.4	14.3	9.3
Start of heading	12.9	7.9	9.8	9.8	9.8	6.4
End of heading	7.6	6.0	6.4	6.0	6.7	4.1
Flowering	6.2	4.3	6.4	4.8	3.6	3.1

A large dose of fertilizers (N_{300} for the summer) makes it possible to cut down slowly aging species (reed canary grass, Timothy grass, meadow fescue) at the age of 50-55 days also.

The dry harvest of clover-rich grass mixtures mowed for the first time, as well as aftermath (not over 40 days old) at the stem-forming and budding stages contained over 8.7 g digestible protein per MJ, and one-half this amount at the start of and during flowering (aftermath 50-70 days old).

Use of large doses of fertilizers would make it possible to perform the first mowing of gramineous grass at the heading stage also, but the experimental results indicate (Table 2) that the grass is rich in protein only at the start of the heading phase, while at the end thereof the level is decreased to two-thirds. In view of the energy to protein ratio, one should not mow gramineous grass later than the start of the heading stage.

It should be recalled that, in most cases, there was more than 10 g protein per MJ metabolic energy at the tillering stage. This means that there is

insufficient energy. It is known that grazing cows are more willing to eat young grass at the tillering stage. The results of the experiments indicate that, at this time, good results are obtained in the form of additional production with a supplement of meal, hay and other energy-rich feed.

When estimating the proportion of energy and protein one must also take into consideration the purpose of the feed. For example, the standard per megalocalorie of metabolic energy is set at 25 g for maintenance feed, 35 g for advancement and 50 g digestible protein for reproduction (6.0, 8.3 and 11.9 g protein per MJ, respectively).

The results of our experiments indicate that, in most cases, the concentration of energy in dry substance of grass feed is consistent with the requirements of a highly productive dairy herd.

The requirements of dairy cows with regard to protein are met by grass feed in the case where the grass is mowed at the booting stage (with a moderate dose of fertilizers) or at the start of the heading stage (with a large dose). For such purposes, clover should be mowed before flowering.

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CSO: 1870

AGROTECHNOLOGY

UDC: 632.93:632.7

PHYSIOLOGICAL AND BIOCHEMICAL EVALUATION OF HARMFULNESS OF GRASS FLIES

Moscow DOKLADY VSESOYUZNOY ORDENA LENINA AKADEMII SEL'SKOKHOZYAYSTVENNYKH NAUK IMENI V. I. LENINA in Russian No 1, 1979 pp 13-14

[Article by P. I. Susidko, Academician of the All-Union Academy of Agricultural Sciences imeni Lenin; V. N. Pisarenko, candidate of biological sciences; and N. Sh. Vygonnaya, All-Union Order of Red Banner of Labor Scientific Research Institute of Corn Growing, submitted 29 Mar 78]

[Text] Investigation of plant indurance of injuries is an important problem in development of complex systems for protecting plants against pests. These systems are based on regulation of number of pests on a specific level, rather than total eradication thereof. For this reason, it is important from the theoretical and practical points of view to study the permissible thresholds of deleteriousness of phytophages.

We conducted our investigations at the experimental base of the All-Union Scientific Research Institute of Corn Growing in 1975-1977.

Various species of grass flies strike winter wheat in the Ukrainian Steppe. Quite often, larvae of several species feed on a single plant. For this reason, we used several species of the order Mayetiola destructor Say., Oscinella frit L. and Phorbia securis T. in our studies. Winter wheat at the 3d stage of organogenesis served as the feed plant. We examined both the above-ground extent of injury to plants and tillering nodes. The intensity of photosynthesis and respiration was determined by the method of L. N. Babushkin. The method of Bertrand as modified by Lisitsin was used to analyze sugar content of tillering nodes of wheat. The obtained data were submitted to mathematical processing.

As a result of these studies, we demonstrated significant deviations in intensity of photosynthesis and respiration of plants injured by larvae of grass flies (Table 1).

There is an increase in intensity of photosynthesis in stricken plants. It reaches maximum levels when the plant's main stem perishes due to injury. In the case of injury to one or two stems, there are less significant changes in intensity. Analogous data were obtained when this index was

determined on plants of the Odesskaya 51 variety. Photosynthesis is closely related to other physiological processes in plants and, first of all, respiration. For this reason, the next stage of our study of the reactions of plants to different levels of damage was to determine the intensity of plant respiration.

Table 1. Change in intensity of photosynthesis and respiration of winter wheat plants (Dneprovskaya 775 variety) stricken by grass fly larvae

Variant	1975	1976	1977
	Intensity of photosynthesis (mg CO_2 dm^2/h)		
Intact (control)	1,98±0,076	2,03±0,173	1,12±0,124
Damaged: central stem	2,36±0,110	2,76±0,112	1,82±0,164
1 lateral stem	2,09±0,131	2,17±0,086	1,46±0,108
2 lateral stems	2,17±0,210	2,22±0,121	1,74±0,186
	Intensity of respiration (mg O_2/g·h)		
Intact (control)	0,412±0,232	0,264±0,117	0,380±0,210
Damaged: central stem	0,846±0,186	0,617±0,205	0,770±0,194
1 lateral stem	0,510±0,194	0,410±0,210	0,616±0,178
2 lateral stems	0,638±0,181	0,486±0,189	0,602±0,106

Table 1 also shows that there is significant increase in intensity of respiration in stricken winter wheat plants. Typically enough, the tendency toward increased intensity of respiration as a function of injury is analogous to that of intensity of photosynthesis. However, there is greater increase in intensity of respiration than intensity of photosynthesis, as a result of which the injured plants lose part of their plastic substances.

The validity of the foregoing is confirmed by the data in Table 2, which lists the results of assaying sugars, the main plastic substances, which play an important role in the process of winter crop wintering. This table shows that there is a significant change in sugar content of tillering nodes of winter wheat when it is damaged. The most significant deviations from the control were noted in plants with damaged main stem or three lateral ones.

However, the reaction of plants of the tested varieties is not the same. Thus, in 1976, in the case of damage to the main stem of Dneprovskaya 775 plants, there was a 14.61% decrease in sugar content, whereas in Odesskaya 51 this index reached only 7.54%. There are negligible changes in sugar content in the case of damage to one lateral stem, and in 1977 plants of the Odesskaya 51 variety of this variant contained even more sugars than the control. Total sugar content in tillering nodes of plants of the above varieties showed negligible deviations in 1976 in the case of destruction of two lateral stems, but in 1977 this index was 55.63% higher in Dneprovskaya 775 than Odesskaya 51. These data are indicative of the greater endurance of the metabolic system of Odesskaya 51 plants when stricken by grass flies.

Table 3. Sugar content of tillering nodes of winter wheat as related to different levels of damage by grass fly larvae

Variant		Dnepr. 775 sugar content (% absol. dry matter) monosacc.	saccharose	total sugar	Odessk. 51 monosacc.	saccharose	total sugar
1976							
Intact (control)		6,76	8,30	15,06	5,69	7,72	13,41
Damaged stems	central	5,34	7,52	12,86	6,11	6,29	12,40
	1 lateral	6,71	8,11	14,82	5,42	7,65	13,10
	2 lateral	6,89	7,92	14,81	6,23	6,58	12,81
	3 lateral	4,51	4,47	8,98	4,40	3,34	7,74
1977							
Intact (control)		6,17	9,23	15,40	6,53	8,71	15,24
Damaged stems	central	6,15	6,60	12,75	5,68	7,33	13,01
	1 lateral	6,46	8,40	14,86	7,00	9,30	16,30
	2 lateral	4,39	7,12	11,51	6,55	8,41	14,96
	3 lateral	3,86	4,78	8,64	5,50	7,41	12,91

Thus, the damage inflicted to winter wheat by grass fly larvae has a significant influence on the nature of synthesis and breakdown of plastic substances in the plant. Odesskaya 51 is more resistant to damage, since it presented considerably less change in sugar content than in the presence of analogous damage to Dneprovskaya 775. Consequently, this is indicative of greater stability of metabolic systems of this variety of plants. The metabolic changes are more significant with damage to the main stem than with damage to even two lateral ones. In the case of injury to one lateral stem, there is insignificant decrease in sugar content, and it is occasionally even higher than in the control, due to activation of compensatory functions of the plant organism.

10,657
CSO: 1870

AGROTECHNOLOGY

UDC: 633.11:631.559

SIGNIFICANCE OF VERTICAL AND HORIZONTAL SYNCHRONIZATION OF PRODUCTIVITY ELEMENTS IN FORMATION OF INTENSIVE TYPE WHEAT HARVEST

Moscow DOKLADY VSESOYUZNOY ORDENA LENINA AKADEMII SEL'SKOKHOZYAYSTVENNYKH NAUK IMENI V. I. LENINA in Russian No 1, 1979 pp 3-5

[Article by Academician V. N. Remeslo; V. F. Sayko, candidate of agricultural sciences; S. N. Zhudra; G. Ye. Borsuk; F. M. Kuperman, doctor of biological sciences; and V. V. Murashev, candidate of biological sciences, Mironovskiy Order of Lenin Scientific Research Institute of Wheat Breeding and Seed Growing and Moscow Order of Lenin and Order of Red Banner of Labor State University imeni M. V. Lomonosov, submitted 9 Nov 78]

[Text] Asynchrony of successive stages of organogenesis is inherent in wild grain species, and this causes significant quantitative variability of development and growth of tillering runners, including significant delay in development of secondary, tertiary and subsequent runners, with corresponding reduction of percentage of their potential productivity.

The property of asynchrony in development of runners [or sprouts] also persisted in grain cultivars, in particular wheat. In most varieties of winter wheat, even with a high coefficient of autumn tillering, constituting 6-7 runners, we observe considerable quantitative differences in growth and rhythm of development of the second, third and subsequent runners, as compared to the first. As shown by the literature [3] and our findings, asynchronous development of tillering runners leads to significant reduction thereof at different stages of organogenesis, different times of formation of reproductive organs of runners and, as a result, a reduced harvest.

It is possible to reduce significantly, by means of specific cultivation technology (in particular, by increasing the sowing norm), the irregularity of maturation of runners of different orders. However, an increase in sowing norm not only leads to unproductive expenditure of seeds, but worsening of grain quality, particularly in years of drought.

In order to reduce the detriment caused by asynchrony of the process of shoot [runner] formation, many breeders are developing wheat varieties with rapidly developing first (main) runners and early reduction of subsequent ones. Wheat

varieties have been developed that form mainly one productive shoot. Under favorable wintering conditions, for example, in Kuban', with high and uniform humidity in the autumn and early spring, varieties such as Bezostaya 1 presented good development and preservation of the planned plant stand density referable to productive runners and good grain content of stalks, i.e., a high and stable harvest [4].

However, in most other regions, the use of cultivars that form one or, less often, two productive shoots cannot assure stable harvests for a number of reasons. For example, during years that have a long and warm autumn, the main shoot progresses to the 3d-4th stages of organogenesis even before the winter, and this often lowers hardiness, causing damage to it. Death of the main shoot is also observed before the snow cover disappears [1]. In such cases, secondary and tertiary shoots can compensate for the loss of the first one and thus provide for a good harvest.

In the last few years, most breeders have tried to develop varieties that form 2-3 productive shoots [6, 7]. This type of bush in highly intensive cultivars increases the harvest significantly as a result of decreased asynchrony of development, growth and productivity of secondary shoots. This is manifested with particular distinction when large doses of fertilizers were used ($N_{120}P_{120}K_{90}$) on the pea precursor, when the number of plants with two shoots increased from 14-30% in the control to 45-64% on a fertilized background in the varieties Mironovskaya 808 and Mironovskaya yubileynaya. Accordingly, there was an increase in participation of second shoots in the harvest (up to 25-30% of total harvest).

High grain content of the spike is an equally important character, as compared to the number of productive shoots, with respect to increasing the yield from cultivars of the intensive type [4, 6]. As we know, in wild wheat species the mean number of grains per spike is 17-20, in wheat cultivars of the extensive type it is 24-27 and in the intensive type, such as Bezostaya 1, Kavkaz, Mironovskaya 808, Mironovskaya 25, Yubileynaya and others, it is 40-55 grains.

Quite recently, in developing new varieties, breeders tried to augment the number of spikelets per spike, i.e., they were concerned with vertical synchronization of spikelet differentiation processes along the axis of the spike at the 3d-4th stages of organogenesis. In a number of cases, this was achieved through interspecific hybridization of wheat with rye, wheat-grass [Agropyron] and wild rye [Elymus], and it was possible to increase the mean number of spikelets to 25-27.

However, only a few varieties (Kavkaz and hybrids thereof, Odesskaya 66) presented a mean number of 22-23 spikelets by the time the spike matured. In most varieties of the intensive type it ranged from 18 to 20, increasing by a mean of 1-2, less often 3 spikelets, with the use of large doses of fertilizer (Table).

Potential and actual productivity of cultivars developed by the Mironovskiy Scientific Research Institute of Wheat Breeding and Seed Growing with the use of different doses of NPK

Variety	Background	Spikelets at stages		Flowers at stages			Caryopsis at 12th st	Harvest (centner/ha)	Productive shoots per m²
		IV	VII	V	VII	IX			
Mironovskaya 808	control	16,8	16,2	123	47	39	32	46,0	325
	$N_{60}P_{60}K_{60}$	17,0	16,6	126	51	47	32	61,1	374
	$N_{120}P_{120}K_{90}$	18,0	17,2	138	51	39	29	66,6	384
Mironovskaya yubileynaya	control	16,0	17,2	123	53	39	31	44,1	380
	$N_{60}P_{60}K_{60}$	18,8	18,2	141	54	46	41	60,5	427
	$N_{120}P_{120}K_{90}$	19,4	19,0	155	58	43	36	66,9	479
Mironovskaya 25	control	17,4	16,2	127	70	40	32	46,4	358
	$N_{60}P_{60}K_{60}$	18,6	17,0	149	75	40	33	60,9	457
	$N_{120}P_{120}K_{90}$	18,7	18,0	152	83	43	32	66,4	464
Mironovskaya nizkoroslaya [short] Il'ichevka	control	17,2	17,1	130	48	37	26	40,6	326
	$N_{60}P_{60}K_{60}$	17,6	17,0	131	51	44	34	54,2	424
	$N_{120}P_{120}K_{90}$	20,0	17,8	164	53	44	32	66,6	485
	control	16,8	16,6	121	53	42	30	40,8	318
	$N_{60}P_{60}K_{60}$	17,4	17,0	136	71	43	35	58,1	496
	$N_{120}P_{120}K_{90}$	18,2	18,2	146	67	47	32	66,4	416

An increase in number of synchronously developing flowers in the spikelets is of much greater importance in increasing grain content per spike [7, 8]. Cultivars of the highly intensive type are notable, not only for potentially large number of flowers at the 5th and 6th stages of organogenesis, but synchronous development thereof at the 7th-8th stages (Figure). When large doses of fertilizer are used, they show a significant increase both in total number of flowers at the 5th stage and in number of caryopsis at the 12th stage, which is particularly inherent in Mironovskaya 25 and Il'ichevka. The new Mironovskiy-bred varieties are appreciably superior to the standard, Mironovskaya 808, in this respect.

As can be seen in the Figure, greater synchrony of flower development in the spikelets at the 7th stage, which we call "horizontal synchronization," yielded 51-42 caryopses in the 1st to 3d runners of the spikes at the 12th stage of organogenesis in a clement year.

In years with radical decline of productive moisture in soil and low relative air humidity, there is reduction of a significant part of the flowers and underdevelopment of caryopses at the 9th to 11th stages, as was the case in 1978. Only a larger number of productive shoots per unit area yielded a significant increase in harvest in the variants with the use of fertilizers (Table).

Thus, morphophysiological analysis of the dynamics of harvest formation in five varieties of winter wheat in 1978, as in a number of other varieties in prior years [2, 9], revealed that synchronized development of productive

shoots at the 2d-3d stages and horizontal synchronization of development of flowers in spikelets, the indicator of which is the number of 3-5-grain spikelets, are of the greatest importance to realization of potential productivity of a variety.

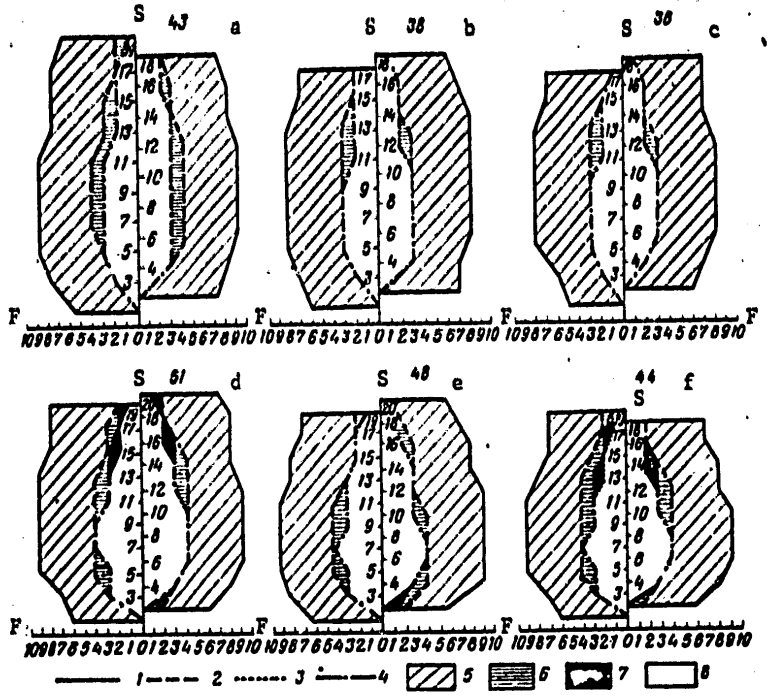

Diagram-models of formation of productivity of spikes of Mironovskaya 808 (a-c) and Mironovskaya 25 (d-f) winter wheat. From left to right: 1st, 2d and 3d shoots; S--spikelets per spike, F--flowers in spikelets. Number of flowers in spikelets and spike:

1) at 5th stage of organogenesis
2) 7th stage
3) 8th-9th stages
4) caryopsis at 11th-12th stages

reduction of spikelets and flowers at:

5) 5th to 7th stages
6) 7th to 8th
7) 8th to 9th
8) grains per spikelets at 10th-12th stages (actual productivity)

The number above each model refers to grain content of spike at 12th stage.

Processes of horizontal synchronization of productivity elements at the 2d-3d and 7th-10th stages of organogenesis are more important to winter wheat than

vertical syncrhonization at the 4th stage, when the number of spikelets per spike is determined. This confirms the importance of the work being done at the Mironovskiy Scientific Research Institute of Wheat Breeding and Seed Growing pertaining to breeding and varietal agrotechnology, aimed at augmenting the number of productivity elements, both by obtaining a tillering coefficient of up to 2.7 for the produtive shoot and increasing the number of caryopses to 60-70 per spike.

In spite of the achieved intensification of the process of horizontal synchronization at the 7th-9th stages of organogenesis, which results in a higher number of caryopses in spikelets of wheat of the intensive type at the next stages, asynchrony of growth processes during formation of caryopses in the spike at the 10th-11th stages of organogesis has not yet been overcome in most varieties. For this reason, continued breeding to overcome asynchronous growth processes and development of appropriate agrotechnical procedures are pressing tasks.

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10,657
CSO: 1870

AGROTECHNOLOGY

UDC: 635.25:575.24

DEVELOPMENTAL DISTINCTIONS OF LEAF SURFACE OF SOME ONION MUTANTS INDUCED BY CHEMICAL MUTAGENS

Moscow DOKLADY VSESOYUZNOY ORDENA LENINA AKADEMII SEL'SKOKHOZYAYSTVENNYKH NAUK IMENI V. I. LENINA in Russian No 1, 1979 pp 19-22

[Article by O. S. Vodyanova, candidate of biological sciences, and K. Tynymbayeva (presented by L. G. Bobrov, corresponding member of the All-Union Agricultural Academy imeni Lenin), Kazakh Scientific Research Institute of Potato and Vegetable Growing, submitted 11 Apr 77]

[Text] Plant breeding is very important in enhancing the effectiveness of photosynthesis. Chemical mutagenesis is one of the methods of obtaining valuable plants with altered photosynthesis [2]. Plants with large leaf area and intensive photosynthesis, capable of maximum formation of spare organs, are promising for southern regions with much light and adequate supply of water.[6].

We conducted experiments in the irrigated region of Alma-Atinskaya Oblast in 1971-1976. Onion mutants isolated in M_2, produced by treating Dungayskiy 56 and Karatal'skiy onion varieties with chemical mutagens--ethylenimine (EI) and N-nitrosoethylurea (NEM)--served as the material for our study; they are characterized by good development of the leaf system. M_1, M_2 and M_3 mutants were planted in families and M_4, in two replicas over an area of 5 m^2, with annual evaluation and examination of the growing generations. Superior grade plants of the same varieties served as a control.

The method of A. A. Nichiporovich [3, 4], modified to be used for onions, served to determine the area of leaf surface and ratio of weight of economically valuable organs to area of leaf surface.

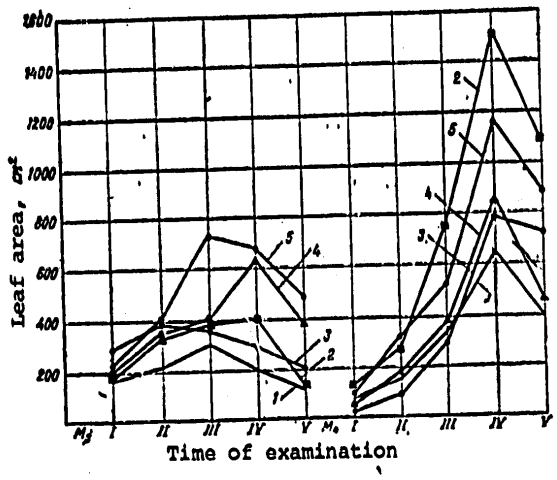
The area of the leaf surface was determined using the formula, $S = (abnK_e) \cdot 2$, where a is the length of the leaf, b is its width and n is the mean number of leaves; K_e is the adjustment coefficient. K_e was established on the basis of 200 measurements of leaves and scapes [flower stalks], and it constitutes 0.73 for leaves and 0.49 for scapes.

Table 1. Leaf surface of mutants of M and M vegetative generations at the time of maximum growth (mean per plant, M±m)

Catalog No of mutant	Chemical mutagen (dosage, %)	Leaves			Leaf area		
		number	length (cm)	width (cm)	cm^2	% of control	Coefficient of variation (Cv, %)
		M_3 Karatal'skiy					
	Control	$\frac{6,4 \pm 0,32}{5,0}$	$\frac{30,0 \pm 1,2}{1,3}$	$\frac{1,1 \pm 0,05}{4,5}$	$\frac{308 \pm 20,2}{7,3}$	100,0	19,9
15-1	NEM (0.05)	$\frac{6,0 \pm 0,2}{3,3}$	$\frac{32,3 \pm 10,9}{3,0}$	$\frac{1,4 \pm 0,05}{3,6}$	$\frac{390 \pm 19,8}{5,0}$	128,5	15,2
15-4	Same	$\frac{10,0 \pm 0,3}{3,0}$	$\frac{34,8 \pm 1,1}{3,4}$	$\frac{1,3 \pm 0,07}{4,9}$	$\frac{661 \pm 45,3}{6,9}$	214,6	15,3
15-7	Same	$\frac{9,0 \pm 0,3}{3,3}$	$\frac{32,0 \pm 1,4}{4,4}$	$\frac{0,9 \pm 0,04}{4,4}$	$\frac{378 \pm 6,6}{1,7}$	122,4	4,0
14-17	EI (0.03)	$\frac{8,0 \pm 0,37}{4,2}$	$\frac{39,0 \pm 0,44}{1,1}$	$\frac{1,6 \pm 0,07}{4,4}$	$\frac{721 \pm 89,5}{8,2}$	234,4	22,9
		Dunganskiy 56					
	Control	$\frac{6,0 \pm 0,44}{7,3}$	$\frac{28,0 \pm 0,089}{3,2}$	$\frac{1,0 \pm 0,03}{3,0}$	$\frac{245 \pm 22,3}{9,1}$	100	
19-5	EI (0.02)	$\frac{5,4 \pm 0,2}{3,6}$	$\frac{32,6 \pm 1,03}{3,3}$	$\frac{1,3 \pm 0,04}{3,0}$	$\frac{234 \pm 26,0}{7,8}$	132,.	
		M_4 Karatal'skiy					
	Control	$\frac{8,7 \pm 0,08}{0,9}$	$\frac{37,8 \pm 2,01}{5,3}$	$\frac{1,2 \pm 0,07}{5,8}$	$\frac{576 \pm 60,0}{10,4}$	100,0	31,2
15-1	NEM (0.05)	$\frac{14,8 \pm 1,18}{7,9}$	$\frac{45,1 \pm 1,7}{3,7}$	$\frac{1,8 \pm 0,08}{4,7}$	$\frac{1530 \pm 64,4}{4,2}$	265,6	37,0
15-4	Same	$\frac{9,0 \pm 0,5}{5,6}$	$\frac{43,2 \pm 0,88}{2,0}$	$\frac{1,7 \pm 0,08}{4,7}$	$\frac{965 \pm 88,6}{9,2}$	167,5	29,0
15-7	Same	$\frac{10,0 \pm 0,34}{3,4}$	$\frac{43,2 \pm 1,0}{2,5}$	$\frac{1,3 \pm 0,01}{0,7}$	$\frac{820 \pm 34,6}{4,2}$	142,4	10,1
14-17	EI (0.03)	$\frac{12,4 \pm 0,79}{6,4}$	$\frac{41,1 \pm 1,3}{3,2}$	$\frac{1,6 \pm 0,07}{4,2}$	$\frac{1191 \pm 86,2}{7,2}$	206,8	22,9
		Dunganskiy 56					
	Control	$\frac{7,8 \pm 0,56}{7,1}$	$\frac{39,9 \pm 1,0}{2,5}$	$\frac{1,2 \pm 0,05}{4,1}$	$\frac{545 \pm 50,8}{9,3}$	100	29,4
19-5	EI (0.02)	$\frac{11,9 \pm 1,1}{9,2}$	$\frac{36,2 \pm 1,02}{2,8}$	$\frac{1,2 \pm 0,04}{0,33}$	$\frac{755 \pm 101,0}{13,4}$	138,5	42,6

Note: Arithmetic mean ($\bar{x}$) and error thereof ($S_{\bar{x}}$) given in numerator and accuracy of experiment (S_v) in denominator.

Biometric measurements were taken of 10 plants in each studied version, with consideration of number of leaves, their length and width, every 10 days until there was visible decrease in leaf surface, which coincides with the time of drying of the lower leaves and start of maturation of the bulb. The experimental data were submitted to mathematical processing according to P. F. Rokitskiy [5] and A. I. Fedorov [7].



Dynamics of leaf surface (mean per plant) in mutants and original Karatal'skiy variety as related to time of examination

For M_3: I) 5 June
II) 15 June
III) 25 June
IV) 5 July
V) 15 July

For M_4: I) 25 June
II) 5 July
III) 15 July
IV) 25 July
V) 5 August

1) Karatal'skiy
2) mutant 15-1
3) mutant 15-7
4) mutant 15-4
5) mutant 14-17

Determination of maximum growth of leaves on the plant and their size, as well as development of area of leaf surface during this period of vegetation, constitute an important indicator of photosynthetic activity of plants. Table 1 lists data on number of leaves per plant, their length and width during the period of maximum growth in third and fourth vegetative generations of mutants.

Table 1 shows that M_3 mutants differ appreciably from their original forms in parameters of leaves and overall leaf surface area. Thus, during the period of maximum growth, the leaf surface area was 22.4-134.% larger in mutants isolated after treating onions with 0.05% NEM and 0.03% EI than in Karatal'skiy. Mutants 15-4 and 14-17 presented a particularly well-developed leaf surface.

The leaf surface area of mutant 19-5 of the Dunganskiy 56 variety, isolated after treating onions with 0.02%, was 32.2% larger than in the control. The coefficient of variation (Cv) of leaf area is lower in M_3 mutants than in the original varieties (the only exception being mutant 14-17, in which the

variability of this character was higher than in the control). In the second year of life (M_3 generative generation) the onion mutants excelled the initial forms by 11.5-108% with regard to overall area of assimilation surface of leaves and scapes.

Fourth vegetative generation mutants (M_4) also differed from the original varieties in leaf parameters and considerably larger area thereof as a whole. The leaf surface area increased by 42.4-165.6% during the period of maximum growth in mutants of Karatal'skiy and by 38.5% in the Dungayskiy 56 mutant, as compared to the control. M_4 mutants 15-1 and 19-5 were notable for greater variability of plants, which caused an increase in the coefficient of variation.

Not only the leaf surface area, but duration of its function, as well as prompt efflux of nutrients to the onions, are of decisive significance to harvest formation.

Table 2. Harvest and ratio of economically useful organs to leaf surface area in third and fourth vegetative generation mutants

Catalog No of mutant	Chemical mutagen (dose, %)	Harvest of commercial onions				Ratio of useful organs to area of leaves over vegetation period			
		M_3		M_4		M_3		M_4	
		kg/м²	% of control	kg/м²	% of control	g/м²	% of control	g/м²	% of control
		Karatal'skiy							
	Control	1,649	100,0	3,167	100,0	1200	100,0	2000	100,0
15-1	NEM (0.05)	2,762	166,8	5,33	153,7	2100	175,0	1700	85,0
15-4	Same	3,756	227,2	8,577	247,3	2300	191,6	4900	245,0
15-7	Same	3,065	185,9	4,32	124,6	2300	191,6	2400	120,0
14-17	EI (0.03)	4,051	245,7	5,22	150,6	1700	141,6	2000	100,0
		Dunganskiy 56							
	Control	1,43	100,0	3,523	100,0	1500	100,0	2600	100,0
19-6	EI (0.02)	2,222	155,2	4,791	135,9	2000	133,3	2900	111,5

The Figure illustrates the dynamics of leaf surface from the time of appearance of the fifth leaf to visible reduction of leaves in our study of M_3 in 1974 and M_4 in 1976. Maximum leaf surface development was observed at the 3d and 5th examination times. In 1974, this coincided with the third 10 days of June and first 10 days of July, and in 1976 with the second and third 10-day periods of July. The differences in time of examining M_3 and M_4 are attributable to the late and cold spring of 1976 [1].

Leaves continue to build up in M_3 for 30 days. M_4 mutants increased their leaf surface for 30-40 days. At the last measurement of leaf area, we observed

a sharp reduction in leaf area: by 27.2-64.6% in M_3 mutants and by 12.2-46.7% in M_4, versus 40-37.3% in the control. There was slower dying off of leaves in mutants 15-7, 14-17 and 15-1, as compared to the control. At the fifth measurement, mutant 15-4 in M_3 and M_4 demonstrated a reduction of leaf surface to over one-half.

In appraising the mutants, special attention was given to demonstration of high-yield variants with a large harvest of commercial onions, as well as to the index of high and stable ratio of weight of important organs to area of leaf surface.

Table 2 shows that the isolated mutants are characterized by a high yield. Thus, in 1974, the M_3 mutants yielded 55.2-145.7% more commercial onions than the control and M_4 yielded 24.6-147.3% more in 1976. The indices of ratio of weight of economically useful organs to leaf area indicate that not all of the mutants make full use of the leaf surface. Mutants 15-4, 15-7 and 19-5 make the most productive use of the leaf surface.

The isolated mutations are of definite interest to breeding work, both for development of green pinna (mutant 15-1) and onions (mutants 15-4, 15-7, 14-17 and 19-6).

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AGROTECHNOLOGY

UDC: 636.5:591.33+591.526

ANALYSIS OF EARLY EMBRYONIC MORTALITY AMONG INTACT, ALEUKOTIC INBRED AND ROUS SARCOMA VIRUS INFECTED CHICKENS

Moscow DOKLADY VSESOYUZNOY ORDENA LENINA AKADEMII SEL'SKOKHOZYAYSTVENNYKH NAUK IMENI V. I. LENINA in Russian No 1, 1979 pp 27-29

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[Text] According to data on the critical periods of embryogenesis of poultry, two or three periods of maximum embryonic mortality are distinguished in poultry farming practice, and it is considered only from the time the eggs are put in incubators. Embryo death occurring in the mother before the eggs are laid, in the so-called oviduct period, is not taken into consideration [6, 7]. Usually, an egg with an embryo that died during this period is classified in the "unfertile" category according to the results of candling. However, studies pursued on mammals have shown convincingly that preimplantation death of embryos, corresponding to fowl death in the oviduct period, constitutes a significant percentage (25-30) of total embryo deaths. I. V. Kudryavtsev [2] discovered, on the basis of his findings in dissection of 11,500 quail eggs, that the index of embryo death in the oviduct period, including unfertilized ovicells, was about 20 times higher than the maximum embryo mortality in the critical period of incubation.

We analyzed the waste from incubation referable to the category of unfertilized eggs after candling, in order to detect eggs with embryos that had died in the oviduct period.

In order to increase the number of unfertilized eggs, we used two factors: inbreeding and Rous sarcoma virus (RSV).

We conducted our work at the Laboratory of Experimental Biological Models of the USSR Academy of Medical Sciences and the experimental farm of the All-Union

Scientific Research Institute of Farm Animal Breeding and Genetics. We included in our analysis aleukotic Russkaya Belaya [Russian White] chickens bred for sensitivity to leukosis with an inbreeding coefficient for the flock of over 35% and autobred fowl of the same breed infected with a measured amount of RSV. Both groups were maintained under the same conditions, the only difference being that the aleukotic chickens were given presterilized feed and water.

The third group consisted of intact autobred White Plymouth Rock chickens. We were aware of the fact that fertilization and hatching rates were somewhat lower in the breeds used for their meat than in laying breeds. The conditions under which these chickens were maintained did not differ from the established standards. In all, we incubated 8058 eggs. The total amount of rejects constituted 2552 eggs. Analysis thereof was made by means of candling and dissection of the eggs. After dissecting unfertilized eggs, they were classified in the cateogry of those that presented no signs of development or arrested development at the oviduct period in the first cleavage divisions, according to degree and nature of development of the embryonic disk (modified Gowe [5] method).

It should be noted that in our evaluation of the data we obtained according to the conventional methods of analysis of incubation results, the parameters of overall waste and components thereof were in the usual permissible ranges for the breed in the group of White Plymouth Rock chickens. At the same time, among the Russkaya Belaya breed, there was more overall waste and part thereof referable to the category of unfertilized eggs. It should be noted that the increase in overall waste and, particularly, percentage of unfertilized eggs in the noninfected group is quite consistent with the findings of other authors who studied the effects of inbreeding on reproductive capacity. It is believed that the significant increase in number of unfertilized eggs is due to the predominant effect of inbreeding on the male, on his reproductive capacity. However, the results we obtained enable us to evaluate differently the effects of inbreeding, if we take into consideration the fact that in the category of unfertilized eggs dissection (Table) can demonstrate that part of the eggs are indeed unfertile, or shows no signs of development, and the other part consists of eggs with embryos that died in the oviduct period.

By means of such more comprehensive analysis, it is only in the Russkaya Belaya aleukotic chickens submitted to the influence of inbreeding that the percentage of really unfertilized eggs remains high, while in those infected with RSV this index is lower than in White Plymouth Rock chickens.

The rest of the unfertilized eggs is referable to early embryonic deaths, probably related to the qualitative characteristics of ovicells or, in other words, contribution of the female to successful development of the embryo.

Results of analysis of incubation rejects

Group	Number of incubated eggs	Rejects, % incubated eggs				
		overall	including			
			with embryo deaths during incubation	"unfertilized"		
				according to candling results	dissection findings	
					with no signs of development	with embryo death in oviduct period
White Plymouth Rock	4640	23.66	11.16	12.50	5.88	6.62
Russkaya Belaya (infected with RSV)	936	32.05	12.71	19.34	4.49	14.85
Russkaya Belaya (aleukotic)	2482	46.49	17.00	29.49	11.32	18.17

It is known, for example, that the first cleavages of the ovicell occur on the basis of using long-lived genetic information which accumulates in the course of oogenesis [1, 3, 4].

In the presence of some defect in one of the genes involved in accumulating such information, the ovicell may be deficient and this, in turn, could lead to embryo death at the very earliest stages of its development, in spite of involvement of normal spermatozoa in fertilization.

The data we obtained on early embryo mortality in the oviduct period of development can be interpreted as the result of qualitative deficiency of ovicells, which is aggravated under the influence of RSV or inbreeding, since embryo deaths are observed at the stage of the first cleavages, which take place without participation of genetic nuclear material and, in particular, male pronucleus. Such qualitative deficiency of the ovicell can occur in the group of aleukotic chickens under the influence of inbreeding, as a result of which homozygous combination of lethal and semilethal maternal genes manifests its action in early embryogenesis, increasing by almost three times embryo mortality in the oviduct period, even as compared to fowl to be used for its meat.

With reference to the correlation between inbreeding and reproductive function of poultry, it must be borne in mind that the index of unfertilized eggs may rise (rather significantly) as a result of embryonic mortality in the oviduct period, due primarily by the influence of inbreeding on the female at the earliest stages of embryo development. In the group of RSV-infected chickens, the high percentage of embryo deaths in the oviduct period can be attributed to the specific interaction between the virus and the mother or direct interaction between the virus and ovicell.

A quantitative comparison of indices of embryo mortality in the oviduct period and during incubation shows that the figures are of about the same order. Embryo death in the oviduct period constituted 37.21% of overall embryo mortality in fowl used for meat, 53.88% in those infected with RSV and 51.66% in aleukotic chickens.

In the course of our studies, we also demonstrated some specific elements in the influence of inbreeding and RSV infection on level and quality of incubation rejects. These specifics consist of the fact that, in the case of inbreeding, the overall waste index and all of its components constituted a rather high percentage, whereas in the group of infected chickens a high percentage was observed only with regard to the index of embryo death in the oviduct period.

Consequently, in implementing biological monitoring of incubation, it is expedient to take into consideration embryo deaths in the oviduct period of development and to use this parameter in evaluating the pedigree qualities of the chickens.

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AGROTECHNOLOGY

UDC: 636.2:591.132.2

INVOLVEMENT OF SOME SUBCELLULAR STRUCTURES AND PROTEINS IN TISSULAR METABOLISM OF RUMINAL MUCOSA

Moscow DOKLADY VSESOYUZNOY ORDENA LENINA AKADEMII SEL'SKOKHOZYAYSTVENNYKH NAUK IMENI V. I. LENINA in Russian No 1, 1979 pp 31-34

[Article by R. M. Shmidt and G. I. Kalachnyuk, doctor of biological sciences (presented by F. Yu. Palfiy, corresponding member of the All-Union Academy of Agricultural Sciences imeni Lenin), Ukrainian Scientific Research Institute of Farm Animal Physiology and Biochemistry and L'vov Zooveterinary Institute, submitted 13 Jun 77]

[Text] In order to accelerate formation of productive qualities in young ruminant animals, it is important to know the processes that occur in the ruminal mucosa, on which the intensity of metabolic processes in the entire organism depends in many respects [4, 5]. Dry mixed feed given to calves from the age of 10-14 days not only aids in earlier formation of the rumen, growth and development thereof, but makes it possible to use carbamide effectively in the calf diet from the 2d month of life on. This effect is probably obtained as a result of intensification of metabolism and biosynthetic processes in nuclear and mitochondrial structures of cells of different tissues and, in particular, of the ruminal mucosa [6].

It was interesting, from the biological and practical points of view, to determine the influence of different forms of carbamide (AKD and nonextruded powdered mixture of components thereof) on indices of protein and nucleic acid metabolism in subcellular structures and hyaloplasm of the ruminal mucosa and other biological objects in calves and fattened bullocks.

For this reason, a new system [7] was used to feed 20 bullock analogues of the black dappled breed dry mixed feed starting at the age of 10-14 days (and starting at 1 month, feed containing carbamide). At 3 months of age, they were given extruded multicomponent supplements containing carbamide (AKD, first group) and the initial mixture used prior to extrusion (loose [powdered] carbamide, second group).

AKD contained coarse corn grain, carbamide, sodium bentonite, sawdust, molasses, phosphatides, feed lard of animal origin, sodium sulfate and santokhin.

The animals were slaughtered at the age of 7 and 14 months. Conventional methods were used to obtain subcellular structures and study their metabolic indices [2-6, 9].

Table 1. Biochemical changes in nuclei of ruminal mucosa and liver of calves given AKD (M±m, n = 4)

Index	Ruminal mucosa		Liver	
	group			
	I	II	I	II
DNA (mg% R[?])	0,50±0,003*	0,38±0,004	2,32±0,035*	1,78±0,015
RNA (mg% R)	1,08±0,007*	0,87±0,008	2,70±0,032*	1,58±0,035
RNA/DNA	2,16	2,31	1,19	0,89
Globulins (mg%)	10,28±0,13*	9,52±0,035	27,26±0,36*	23,09±0,67
Histones (mg%)	17,91±0,54	17,78±0,61	41,93±1,21	35,42±0,85
Acid proteins (mg%)	46,39±0,66	45,00±0,24	118,2±1,24	83,93±1,56
Membrane complex proteins (mg%)	420,0±2,78*	369,8±2,72	189,4±2,97*	160,7±4,51
RNP number (activity units)	11,21	9,26	22,46	16,62
DNP number (activ.un.)	16,66	10,74	14,56	13,70
rRNP, mg%	5,28±0,12	5,11±0,28	30,80±0,60*	16,58±0,76
mRNP (mg%)	1,72±0,11	1,94±0,11	21,03±0,49*	18,51±0,55
oRNP (mg%)	101,85±0,48*	96,30±0,38	42,66±0,50*	35,26±0,50
rRNP number (activ.un.)	20,83	16,05	8,80	12,48
mRNPA number (activ.un.)	58,14	40,21	12,60	13,29
oRNP number (activ.un.)	8,54	7,38	52,13	32,05

* P≤0,05.

Table 1 shows that there was an increase in concentration of RNA and DNA in the ruminal mucosal nuclei of animals given AKD. The RNA/DNA ratio was about the same in both groups of bullocks. Analogous findings were made in liver tissue. The ratio of acid to histone proteins, which are the possible regulators of DNA gene activity, was also the same in both tissues (2.59 and 2.53). However, this ratio was reliably higher in the liver of the first group of animals than the second (2.81 and 2.33, respectively). There was a reliable increase in membrane complex protein in both the rumen and liver. There was twice as much in the ruminal mucosa as in the liver. These proteins are probably bound with the nuclear membrane of the ruminal mucosa and, perhaps, are intended for extraction ["exportation"], since the mucosa synthesizes complex glycoliponucleoprotein complexes mainly for extraction [4].

The increase in RNP number in nuclei of the ruminal mucosa of animals in the first group corresponds to an analogous increase thereof in the liver, but nuclear RNA activity of the latter is twice as high as in the mucosa. The DNP number is 50% higher in calves of the first group. This is related to the fact that there are fewer proteins here, extracted with this nucleic acid fraction, per unit DNA. We failed to demonstrate reliable differences in the liver according to this index. Consequently, the ruminal mucosa is the first to react to a change in quality of diet. Its high rRNP number, as compared to the liver, is also indicative of lower protein content in the rRNP complex of the mucosa. The level thereof is 5-6 times lower than in

the liver, and there is 10 times less protein in mRNA. While rRNA and mRNA activity of the liver is virtually the same, the mRNP number is 2.5 times higher in the ruminal mucosa than the rRNP number, which may be indicative of intensified involvement of mRNA in processes of transmission of the genetic code to the site of protein synthesis. A reliable increase in oRNP, i.e., protein in the fraction of de novo RNA, in the first group of animals, as well as increase in RNP number of this complex should, according to the data of A. N. Belozerskiy [1], be associated with more intensive metabolism. The 5-6-fold increase in oRNP number in the ruminal mucosa, as compared to the liver, is additional evidence of the fact that the mucosa functions for "exportation" [4-6]. The low protein content of DNP and RNP complexes creates, in our opinion, the high metabolic activity of the tested biological objects. The increase in globulin protein fraction in nuclei of the ruminal mucosa and liver of calves in the first group may be indicative of increase in levels of protective agents in the organism.

Table 2. Metabolic activity of subcellular fractions of ruminal mucosa and liver of calves (units of activity in the two groups)

Index	Ruminal mucosa		Liver	
	I	II	I	II
Mitochondria				
RNP number	9,68	8,47	12,11	10,84
DNP number	3,98	4,06	1,06	1,06
Storage number	54,73	44,07	73,66	75,00
Microsomes				
RNP number	10,21	10,19	7,72	6,94
Storage number	63,00	58,34	68,83	73,27
Ribosomes				
RNP number	10,02	11,34	8,92	7,16
Storage number	56,14	58,25	54,43	54,31
Hyaloplasm				
RNP number	6,01	4,80	1,40	1,20
Storage number	2,57	1,97	24,79	26,03

The results of studies of the mitochondrial and ribosomal system are consistent with the above conclusions (Table 2). The protein reserves in the mitochondria (storage number) are significantly higher in the ruminal mucosa of the first group of animals than the second, but somewhat lower than in the liver. Metabolic activity of RNA and protein reserves in microsomes and ribosomes of the ruminal mucosa are higher than in the liver, with the exception of the storage number in microsomes. In the cytosol supernant of mucosa, the RNP number is 4-5 times higher than in the liver, while the protein reserves are very negligible, as compared to the latter.

The proteins of the hyaloplasm of the ruminal mucosa can be separated into 12-14 main fractions by means of disk electrophoresis. Posttransferrins (5 subfractions) are the most heterogeneous. The γ-globulin zone does not have a diffuse background that is inherent in serum proteins. A higher protein content (unlike blood serum) was found in the postalbumin region, which is represented mainly by α-globulins (4 fractions). Their number is reliably higher in the first group of animals (Table 3). Immunophoresis with homologous antiserum showed that soluble proteins of the ruminal mucosa in the α-globulin region form two paired components, which may be indicative of synthesis of additional α-globulins by the ruminal mucosa.

Table 3. Disk electrophoregram of proteins in cytosol supernantant of ruminal mucosa and blood serum of fattened bullocks (% of total protein in the two groups)

Protein fractions	Ruminal mucosa		Liver	
	I	II	I	II
Albumin	4,00±0,13*	6,90±0,03	15,79±0,08*	13,76±0,05
Postalbumins	22,40±0,35*	20,00±0,56	6,50±0,12*	4,13±0,60
Transferrins	16,80±0,12	17,20±0,44	13,39±0,35*	8,80±0,14
Posttransferrins	40,80±0,61	42,10±0,04	35,25±0,08	37,96±0,02
α2 = macroglobulin + β = lipoprotein	16,0±0,01*	13,79±0,75	10,67±0,15*	14,44±0,32

Note: 1st group--animals given AKD; 2d--sunflower grist [or oil cakes]
* P<0,05.

On densitograms, the transferrins of the ruminal mucosa are represented by one peak, and they constitute 16-17% of total protein. Transferrins are characterized by moderate mobility in an electric field. Their levels are lower in blood serum, as compared to the mucosa, but reliably higher in animals given AKD. On the densitograms, α_2-macroglobulin (α_2Mg) and lipoprotein (β-Lp) merge into one peak constituting 13-16% of the total amount. This is somewhat higher than in blood serum. The amounts of these complexes in the ruminal mucosa are reliably larger in the first (experimental) group of animals. Immunologically, we demonstrated that IgM is a specific protein of the ruminal mucosa. A precipitate thereof is not formed with antiserum against blood serum proteins and soluble proteins of the ruminal mucosa, which warrants the belief that it is specific, i.e., it is synthesized in cells of the latter.

Albumin was found in relatively small amounts in the ruminal mucosa (4-7%). This very labile protein, with relatively low molecular weight, passes into the ruminal cavity from blood [8]. We failed to demonstrate by immunoelectrophoresis a line of precipitation thereof among the proteins of the ruminal mucosa against homologous antiserum. Perhaps it is indeed of exogenous origin, or else migrates sooner than other proteins. The reliable decrease in blood albumin content in the second (control) group and concurrent increase in the ruminal mucosa may be indicative of intensive transport thereof from blood or increased synthesis in this tissue.

The results of disk electrophoresis of proteins of the ruminal mucosa and blood serum indicate that feeding AKD to bullocks being fattened affects the proportion of protein fractions, raising the levels of α-globulins, α_2Mg and β-Lp complexes in the mucosa and albumin, transferrin and α-globulins in blood serum, thereby stimulating metabolic processes in the animals.

Thus, the data we obtained are indicative of active involvement of nuclear, mitochondrial, ribosomal, cytoplasmic proteins and nucleoproteins in intertissular metabolism of the ruminal mucosa.

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CSO: 1870

AGROTECHNOLOGY

UDC: 628.322

THEORETICAL SUBSTANTIATION OF THE PROCESS OF THERMAL DECONTAMINATION OF LIQUID SEWAGE

Moscow DOKLADY VSESOYUZNOY ORDENA LENINA AKADEMII SEL'SKOKHOZYAYSTVENNYKH NAUK IMENI V. I. LENINA in Russian No 1, 1979 pp 34-36

[Article by V. A. Kokurin, candidated of engineering sciences; I. A. Bakulov, doctor of veterinary sciences; and V. M. Kotlyarov, candidate of biological sciences (presented by V. S. Yershov, academician of the All-Union Academy of Agricultural Sciences imeni Lenin), All-Union Scientific Research Institute of Veterinary Virology and Microbiology, submitted on 27 Jun 78]

[Text] Drainage water [or sewage] is a multiphase system in which solid and liquid disperse phases dominate. The process of thermal decontamination takes place simultaneously in both phases. In the case of continuous contact in the "liquid-solid" system, the mutual influence of different factors is involved, and this complicates analysis of the mechanism of thermal inactivation of microorganisms.

In view of the fact that the opinions of scientists are contradictory on this score, it is interesting to take a closer look on the physical essence of the process of heat decontamination of sewage contaminated with microorganisms.

Thermal inactivation of microorganisms in a wet environment is a complex process that depends on a number of physical, biochemical and other factors. This process has been the subject of numerous investigations. However, there is still no precise explanation of phenomena of heat-resistance of microorganisms and causes of inactivation thereof [1-6].

There are typical cases of inactivation in a more complex two-phase "liquid-solid" system. Of course, in the presence of a solid phase, the main process is even more complex and proceeds in many stages. One of the chief elements in the process is diffusion of inactivating agent toward the microorganism (third stage) by means of using a heat carrier (steam) in the medium to be treated. The heat balance equation can be used to determine the patterns of change in temperature during sterilization of liquid with live steam in periodic action apparatus.

Disregarding the negligible expenditure of heat to warm the air space above the surface of the liquid and heat loss in the ambient environment, the heat balance equation for the time of warming of the liquid can be written down as follows:

$$M \cdot d\tau (i_0 - C_{\text{ж}} \cdot t) = V_1 \rho_1 \cdot C_{\text{ж}} \cdot dt + M_{\text{к}} \cdot C_{\text{к}} \cdot dt \quad (1)$$

where M is mass expenditure of heat carrier (steam) through the bubbler (kg/s); τ is time of contact (s); i_0 is specific enthalpy of steam entering the tank; $C_{\text{ж}}$ is thermal capacity of liquid (kJ/kg×deg); t is temperature of the liquid ($^{\circ}$C); V_1 is the volume of liquid in the apparatus (m^3) and ρ_1 is the density of the liquid (kg/m^3);

$$M_{\text{к}} = \sum_{i=1}^{n} m_i \cdot C_i$$

is the sum of products of masses of different parts of the apparatus, $C_{\text{к}}$ is thermal capacity of the apparatus material (kJ/kg·deg).

In the opinion of Bachrach [5], when microorganisms and, in particular, viruses are treated with heat, the hydrogen bonds break and there is disruption of spatial mutual location of structural components of its capsid. As a result, the virus loses its initial functions. According to the experiments of Lauffer and Price [6], inactivation at high temperatures occurs under the influence of two factors: break of hydrogen bonds and denaturation of proteins.

The decontamination process proceeds through the following main stages: I--concentrated delivery of inactivating agent; II--turbulent mixing of fluid; III--diffusion of inactivating agent toward microorganism; IV--penetration of inactivating agent through the membrane of a live cell; V--action of inactivating agent on live cell plasma; VI--dying of microorganism.

In the presence of a solid disperse phase in the liquid medium, the former can have a protective action, preventing penetration of inactivating agent to microorganisms within the solid particles. In this case, the 4th stage consists of two substages: first penetration into the liquid medium to the particles and then through the solid membrane of the particles to the microorganisms.

The amount of heat transferred to the liquid in the course of inactivation of microorganisms is used to raise the temperature from t_{in} to $t_{\text{ж}}+dt$ in the fluid tank and body of the apparatus. In this case, the equation of heat balance (1) can be converted to a more convenient formula to define the patterns of change in temperature of the liquid.

For this purpose, let us divide both parts of the equation by $d\tau$ and transfer all terms to the left, after which we shall obtain:

$$\frac{dt}{d\tau} + \frac{M \cdot C_M}{V_1 \cdot P_1 \cdot C_M + M_R \cdot C_R} \cdot t - \frac{M \cdot t_0}{V_1 \cdot P_1 \cdot C_M + M_R \cdot C_R} = 0, \quad (2)$$

$$\frac{dt}{d\tau} + A \cdot t + B = 0, \quad (3)$$

$$A = \frac{M \cdot C_M}{V_1 \cdot P_1 \cdot C_M + M_R + C_R};$$

$$B = \frac{M \cdot t_0}{V_1 \cdot P_1 \cdot C_M + M_R \cdot C_R}.$$

Equation (3) can be classified as a first order differential equation. With constant expenditure of steam during heating of liquid, parameters A and B are constant.

In order to derive the general pattern of change in temperature, we first solve the following homogeneous equation:

$$\frac{dt}{d} + A \cdot t = 0 \quad (4)$$

Integrating (4) in the range of 0 to t, we shall have:

$$\text{In}t = -A\tau + \text{In}K, \quad (5)$$

where K is the arbitrary integration constant.

According to equation (5):

$$t = K \cdot e \quad (6)$$

We find the general solution for (3) by the method of variation of the arbitrary constant (4):

$$\frac{dt}{d\tau} = \frac{dK}{d\tau} \cdot e^{-A\tau} + K(-A) \cdot e^{-A\tau}. \quad (7)$$

After substituting (6) and (7) in (3) and certain transformations, we shall have:

$$\frac{dK}{d\tau} \cdot e^{-A\tau} + K(-A) \cdot e^{-A\tau} + A \cdot K \cdot e^{-A\tau} + B = 0. \quad (8)$$

Since $\frac{d\kappa}{d\tau} + B \cdot e^{A\tau} = 0$,

$$d\kappa = -B \cdot e^{A\tau} \cdot d\tau. \quad (9)$$

and

$$\kappa = \kappa_1 - \frac{B}{A} \cdot e^{A\tau}. \quad (10)$$

Substituting (10) in (6) we have: $t = \kappa_1 \cdot e^{-A\tau} - \frac{B}{A}$. (11)

Equation (11) is the general solution for (3).

At the start of the process ($\tau = 0$), the temperature in the apparatus equals t. In this case, we have:

$$\kappa = t_1 + \frac{B}{A}; \quad (12)$$

$$t = \left(t_1 + \frac{B}{A}\right) \cdot e^{-A\tau} - \frac{B}{A}. \quad (13)$$

Substituting the values of A and B in (13), we shall obtain the general formula for change in temperature of inactivation in the liquid phase:

$$t = \left(t_1 - \frac{i_0}{C_{\text{ж}}}\right) \cdot e^{-\frac{M \cdot C}{V_1 \cdot \rho_1 \cdot C_{\text{ж}} + M_{\text{к}} \cdot C_{\text{к}}}} + \frac{i_0}{C_{\text{ж}}}. \quad (14)$$

Equation (14) describes the kinetics of heating the liquid to the inactivating temperature. According to (14):

$$\frac{i_{\text{к}} - \frac{i_0}{C_{\text{ж}}}}{t_1 - \frac{i_0}{C_{\text{ж}}}} = e^{-\frac{M \cdot C_{\text{ж}}}{V_1 \cdot \rho_1 \cdot C_{\text{ж}} + M_{\text{к}} \cdot C_{\text{к}}} \times \tau}. \quad (15)$$

Solving (15) for τ, we have:

$$\tau = \frac{V_1 \cdot \rho_1 \cdot C_{\text{ж}} + M_{\text{к}} \cdot C_{\text{к}}}{M \cdot C_{\text{ж}}} \times \ln \frac{t_1 \cdot C_{\text{ж}} - i_0}{t_{\text{к}} \cdot C_{\text{ж}} - i_0}. \quad (16)$$

The obtained equation (16) enables us to determine in general the total time required to heat the liquid to the temperature of inactivation of pathogenic microorganisms it contains.

The data obtained as a result of experiments served as the basis for designing an apparatus for decontaminating liquid sewage by the thermal method to inactivate vegetative and sporulated forms of microorganisms. This device is being introduced on a broad basis to enterprises of the microbiological industry and scientific research institutes, as well as for decontamination of manure at livestock complexes, in particular at the 54,000-head pig complexes, Vladimirskiy and Suzdal'skiy.

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CSO: 1870

AGROTECHNOLOGY

UDC: 636.4:636.084.49

COMPARATIVE EVALUATION OF THE SEDATIVE EFFECTS OF DIFFERENT TRANQUILIZERS ON WEANED PIGLETS

Moscow DOKLADY VSESOYUZNOY ORDENA LENINA AKADEMII SEL'SKOKHOZYAYSTVENNYKH NAUK IMENI V. I. LENINA in Russian No 1, 1979 pp 38-40

[Article by M. N. Kuvichkin (presented by P. Ye. Ladan, academician of the All-Union Academy of Agricultural Sciences imeni Lenin), Don Order of Red Banner of Labor Agricultural Institute, submitted 9 Mar 78]

[Text] In view of the increasing demand by the public for high-quality lean pork, it is necessary to develop specialized pork breeds and strains in the swine breeding industry. It has been established that there is a decrease in swine resistance to stress factors with improvement of pork qualities [1-4]. Such animals are characterized by hormonal and vegeto-nervous instability, increased sensitivity of the cardiovascular system, unsatisfactory oxygen transport by blood, limited heat regulation, etc. [5-8].

We investigated the effect of the stress factor of weaning on physiological parameters of piglets, morphological and biochemical properties of blood, and we also made a comparative evaluation of the sedative effects of different tranquilizers given to piglets when they are weaned, in order to determine which were the most effective.

The studies were conducted on Rostov pork pigs bred at the Don Agricultural Institute by crossing Large White, White Short-Eared, P'yetren and Wales breeds, followed by selection of the obtained hybrids. We took 2-month piglets, of analogous weight and development, in our experiments. Their diet was consistent with the VIZh [All-Union Scientific Research Institute of Livestock Breeding] norms. We formed seven groups of animals (Table 1) from the total number of litters.

An aminazine [thorazine] injection was given 1 day before weaning and for 3 days thereafter, twice a day (in the morning and evening); the other agents were given in lozenge and tablet form three times (morning, lunchtime and evening). Blood was taken for analysis 3 days before weaning, on the 2d, 7th and 15th days after. We counted erythrocytes, leukocytes, hemoglobin, eosinophils per mm^3 blood, total protein, protein fractions, total cholesterol,

glutamic-aspartic and glutamic-alanine transaminases (AST and ALT), and we determined the leukocyte formula. At the same times, we measured body temperature, pulse and respiration rate, and studied the behavioral reactions of the piglets.

Table 1. Experimental set-up (3 litters per group, 4 piglets per litter)

Group	Agent and form thereof	Route of administration and dosage (mg/kg weight)
I control		
II	Aminazin (2.5% solution)	Intramuscular (1.0)
III	Aminazin (lozenges)	By mouth (1.5)
IV	Triftazin [stelazine] (tablets)	" " (1.0)
V	Oxylidin (tablets)	" " (1.5)
VI	Majeptil (tablets)	" " (0.15)
VII	Frenolon [methophazine] (tablets)	(0.30)

In the preweaning period, the parameters studied were in the normal range in all groups of piglets. The weaning period did not affect the number of erythrocytes, hemoglobin, total protein and protein fractions. On the 2d day after weaning, body temperature was 0.7°C higher, pulse rate was 28 beats/min faster ($P<0.01$) and respiratory rate 21 excursions/min faster ($P = 0.001$) in the control group of animals, as compared to the preweaning period. Eosinophil count per mm^3 blood dropped by 43% ($P<0.001$). These indices reached normal levels only on the 15th postweaning day. Analogous results were obtained after giving oxylidin.

Intramuscular and oral administration of aminazine, as well as triftazin and frenolon, was associated with a negligible drop of body temperature, pulse and respiration rate, as well as eosinophils per mm^3 blood in the postweaning period. However, these changes were within the permissible range.

With the use of majeptil, body temperature dropped by 1.2°C, pulse rate by 44 beats/min ($P<0.001$) and respiration rate by 16 excursions/min ($P<0.001$) on the 2d postweaning day, as compared to the preweaning period. These parameters were consistent with the norm only after 15 days.

Analysis of the leukocyte formula before and after weaning the piglets revealed that weaning without the use of tranquilizers and with administration of oxylidin was associated with a 1.2% decrease in eosinophils ($P<0.01$), and the level thereof reached that of the preweaning period only after 15 days; this occurred by the 7th day with the use of the other agents.

The levels of protein fractions were the same before and after weaning in all groups: Thus, albumins constituted 41.37-42.49%, α-globulins 20.69-21.70%, β-globulins 13.86-15.83% and γ-globulins 21.76-22.47%.

Table 2. Biochemical parameters of piglet blood (M±m)

Group	Total protein (g%)	Transaminases (units/mℓ) AST	ALT	Total cholesterol (mg%)
I (control)	6.79±0.19	49.56±2.63	45.28±2.43	118.63±5.31
	6.67±0.18	58.23±2.87	52.02±2.67	102.18±4.47
	6.12±0.14	57.18±2.72	51.26±2.51	106.26±4.40
	6.56±0.15	49.48±2.43	44.96±2.37	117.88±5.12
II	6.48±0.13	48.36±2.34	44.97±2.38	117.26±5.10
	6.39±0.11	49.04±2.40	45.17±2.26	116.76±4.93
	6.27±0.09	49.23±2.51	45.53±2.28	116.94±4.64
	6.43±0.12	48.77±2.37	45.03±2.18	116.98±4.87
III	6.63±0.17	49.12±2.12	45.16±2.24	116.72±3.93
	6.28±0.09	49.96±2.13	46.07±2.31	114.34±3.78
	6.34±0.10	50.10±2.47	45.73±2.23	115.37±3.83
	6.58±0.16	49.38±2.16	45.38±2.18	115.96±3.90
IV	6.54±0.14	48.73±2.30	44.53±2.36	117.83±4.73
	6.18±0.06	49.14±2.37	45.17±2.29	116.96±4.14
	6.27±0.08	49.27±2.34	45.02±2.18	115.82±4.02
	6.43±0.13	48.86±2.24	44.76±2.13	117.29±4.69
V	6.67±0.17	49.11±2.15	45.17±2.31	116.86±4.11
	6.48±0.13	56.94±2.74	51.03±2.34	98.96±3.17
	6.12±0.04	55.73±2.68	47.04±2.23	103.14±2.71
	6.39±0.12	48.43±2.17	45.53±2.27	115.94±4.72
VI	6.74±0.17	48.86±2.20	44.73±2.18	118.02±5.17
	6.12±0.04	49.94±2.26	45.12±2.31	117.16±4.73
	6.17±0.05	48.37±2.18	45.03±2.13	116.84±4.56
	6.27±0.07	48.85±2.17	44.96±2.24	117.83±4.80
VII	6.99 0.18	48.12±2.19	44.85±2.27	116.43±4.03
	6.47 0.12	49.37±2.31	45.32±2.37	115.87±3.90
	6.72 0.16	48.76±2.30	45.03±2.20	115.98±3.94
	6.54 0.13	48.27±2.81	45.88±2.87	116.67±3.80

Note: The first line refers to 3 days before weaning, the second to fourth lines refer to 2d, 7th and 15th days, respectively, after weaning.

Table 2 shows that there was an increase in AST activity by 8.67 units/mℓ and in ALT activity by 6.74 units/mℓ ($P>0.95$) on the 2d day after weaning in the control group of animals. Total cholesterol decreased by 16.45 mg%, as compared to the preweaning period ($P<0.01$). These parameters reached the preweaning levels 15 days after weaning. Analogous results were obtained with the use of oxylidin. The other agents stabilized the biochemical indices of blood at one level.

A study of behavioral reactions of the piglets revealed that they spent 24.3% of the day on eating feed and suckling, 21.4% in active behavior, 21.0% lying down and 33.3% sleeping. In the postweaning period, restlessness was observed in the first and fifth groups of animals; they ate feed unwillingly and in small quantities; they moved about more and rested less. A smaller part of the day was spent lying down and sleeping than before weaning, the decrease constituting 4.6 and 3.7%, respectively. Majeptil depressed the animals. They refused to eat and spent 79% of the day lying down and sleeping. The other agents did not have an appreciable effect on the animals' behavior. They were relaxed and spent 20.8, 19.2, 21.7 and 38.3% of the day eating feed, moving actively, lying down and sleeping, respectively.

Thus, our studies failed to demonstrate removal of the adverse effect of the stress factor of weaning by oxylidin, and we demonstrated a depressing effect on piglets of majeptil, when given to the piglets in the above-indicated doses at the age of 2 months. Administration of 2.5% solution of aminazin intramuscularly in a dosage of 1.0 mg/kg body weight, aminazin, triftazin and frenolon by mouth in doses of 1.5, 1.0 and 0.30 mg/kg, respectively, stabilized physiological processes, leading rather rapidly to normalization of existing deviations of physiological and biochemical indices.

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CSO: 1870

AGROTECHNOLOGY

UDC 631.81:681.3

COMPUTER INFORMATION INPUT HAVING DATA ANALYSIS AND ERROR CORRECTION FEATURES

Moscow DOKLADY VSESOYUZNOY ORDENA LENINA AKADEMII SEL'SKOKHOZYAYSTVENNYKH NAUK IMENI V. I. LENINA in Russian No 3, 1979 pp 34-36

[Article by Cand Tech Sci R. A. Srapenyants, M. M. Mkrtchyan, and L. V. Arutyunova, All-Union Order of the Red Labor Banner Scientific Research Institute of Fertilizers and Agricultural Soil Science imeni D..N. Pryanishnikov]

[Text] The VIUA [All-Union Scientific Research Institute of Fertilizers and Agricultural Soil Science imeni D. N. Pryanishnikov] has created an automated (computer-based) system for collecting, storing, processing, and generalizing data from field fertilizer experiments on a countrywide scale, given the code name "VIUA Geographical Network Agrochemist." This system embodies a number of original concepts which may be utilized to mechanize and automate the processing of scientific research data, and in support of agricultural automated control systems.

The general-purpose information and reference system (data bank) created at the VIUA is described below; it can be used as the basis for organizing data banks for various information systems.

A general-purpose information and reference system (1,3) must satisfy the following input program requirements: Universality; the possibility for introducing packages of input information prepared automatically; acceptance of input from various devices; presence of a special editing language; maximum utilization of standard software in the system.

In addition the method by which data are prepared for input plays a significant role in the organization of an information bank (2,4). Inasmuch as information preparation is as a rule the most laborious process, one which does not readily yield to automation, its correct organization would significantly reduce the time required to prepare input information, decrease the number of possible errors, and facilitate the operator's work. We know that the success with which the organizational structure of information fed into a computer is chosen depends on the correctness with which the input information is prepared.

Let us examine the organization of an input system developed for the information data bank for the "VIUA Geographical Network Agrochemist" Information and Reference System.

Preliminary information preparation: The units of input information are data from one experiment on a special report form. The information is recorded in a formalized format convenient for translation to computer carriers (punchcards, punched tape).

The units of information on each report are text of varying lengths. Therefore it would be suitable to substitute them by identifiers of standard length, which would economize on the memory and simplify the structure of the system, reduce the volume of edited data, and increase the effectiveness of information retrieval. For these purposes we often employ external coding making use of special codes applied to words during preliminary data preparation.

The unique features of the system (large, constantly increasing volumes of information, the possibility for introducing new input data formats, the need for obtaining information in a form convenient to reading) require the use of internal coding. For this purpose we created the Thesaurus (2,4) (automatic identifier dictionary). The dictionary is recorded on a disk-type memory and is easily enlarged.

Information automatically translated into computer carriers is then fed into the computer in the form of data packages.

The input subsystem (the complex of programs supporting information input) is usually in computer codes (1,3,4). This coding is relatively simple, and ensures a high information input rate, which has significance to systems processing a continuous information flow.

In our information and reference system the information is fed into the bank passively--that is, it is introduced into the computer as packages initially prepared in autonomic mode. Because the program has to be reproduced for computers of other types, it was suitable to write the input program in a high-level language. The insignificant decline in input rate does not significantly affect massive information input.

External representation of information: The subsystem's input unit is a report consisting of natural or coded responses to all or some (out of 192) questions stated in a format determined outside the subsystem. The report's questions differ in relation to types and methods of representation, as well as in relation to the quantity of elementary data in the response, and they are subdivided into the following groups: Integer; fraction 1--a real number introduced and stored with a precision of 0.1; fraction 2--a real number introduced and stored with a precision of up to 0.01; vector--a pair or natural numbers, for example data taking the form of days and months, with the division point mandatorily indicated; text c--continuous text of

indefinite length without division points; text ϕ--continuous text of fixed length.

For considerations of economic use of the disk-type memory, integral variables are chosen as the means of internal data representation.

The number of responses to questions contained in the report or the dimensionality of the characteristics is documented outside the system, or it is specified by the value of characteristics N_B (number of experimental variants) and N_Π (the number of experimental replications).

The questions are subdivided into several groups depending on dimensionality (the number of responses is fixed, the number of responses is N_B, the number of responses is unpredictable, and so on).

Serial numbers defining the input formats in the format generator are assigned to the types of questions listed above.

Internal representation of the report: The retrieval requirements, which were determined outside the system, necessitated a report consisting of a line of nine blocks. The first block contains a description of the locations of the responses to the questions, with a consideration for the possibility that some fields of the report would not be filled in. The remaining blocks contain responses to the questions of the report, condensed to eliminate empty spaces with the goal of economizing on carrier volume.

Control tables of the input oscillator, and its tuning: During information input the subsystem analyzes the incoming questions and responses, typing information on errors, if they exist, and discarding incorrect data.

Such analysis is performed with the help of the input oscillator's control table, which stores two parameters for each of N questions: T--type of question, and O--number of responses.

The oscillator is tuned with a special directive; three parameters of the directive prescribe, correspondingly, the number of the question to be inspected, the question type value, T, and together with it the number of questions O, if it is required by context.

The carrier format and the data preparation rules: Either punched tapes or punchcards in standard code are used as the report carrier. During input, the oscillator monitors correspondence of the type and quantity of the introduced data to the specifications recorded in the tables described above. Such monitoring is impossible without introduction of some sort of divider.

Semantic switch-on of the divider is foreseen in the subsystem. The divider marks the beginning of the number of each new question for which the answer follows. Any symbol not used in the data can be adopted as the division symbol. An additional divider was adopted to mark the end of a report.

The comma symbol (,) was adopted to separate elements of the numerical information. For convenience of data preparation, use of any quantity of empty spaces desired between two successive units of information is permitted.

The oscillator also has a table of symbols equated to empty spaces in the input of numerical information; presence of such a table makes it possible, during the carrier's preparation, to use symbols for things such as carriage return and line shift, which is often found to be convenient. This table can be altered by means of separate special communication with the oscillator's service module, similarly as with alteration of control tables.

The procedure of information input, syntactic analysis, and storage: The first input operation is read-out of information contained in the report, from the selected input device to an intermediate file on a disk. The input routine subsequently makes use of this file. This structure makes it possible to correct all errors discovered by the input program with the assistance of standard computer software for text editing. Such software is available for all modern computers; it uses a well developed language that is adapted to the structure of the given computer and has been fully developed. This concurrently precludes the need for the authors of the reports to write special software with which to edit the input reports. The input program can only search for and classify the errors. It communicates the types of such errors and the places where they are discovered to the operator.

Information on each of the reports that are recorded in the file is read into the internal memory in batches, and it is subjected to syntactic analysis.

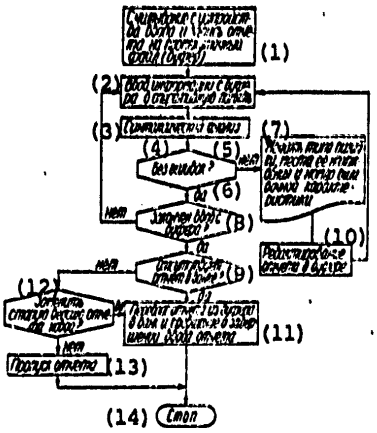
Syntactic analysis is performed in the following order: Analysis of the current question being fed in (communication with the format oscillator and establishment of the type and dimensionality of the question); analysis of data in relation to the type of current question (checking the question type and communicating presence of errors, if they exist); analysis of a question in relation to its dimensionality (checking the dimensionality, and communicating errors).

In addition to the checks listed above, the program is responsible for analysis of the controlling part of the report (presence of unique attributes--year and serial number, mandatory questions--the number of variants and replications).

All error reports are printed out, and they have the form:

Text 1
a, b, Text 2,

where Text 1 is the cell content (a precise copy of the carrier in the place immediately preceding the error), a is the serial number of the last



Input Subsystem Block Diagram

Key:

1. Readout from input device and recording of report on intermediate file (buffer)
2. Information input from buffer into internal memory
3. Syntactic analysis
4. No errors?
5. No
6. Yes
7. Printout of type of error, its location, and serial number of wrong characteristic
8. Input from buffer complete?
9. Report absent from bank?
10. Editing of report in buffer
11. Transfer of report from buffer to bank, and communication of signal indicating completion of report input
12. Replace old version of report by new?
13. Pass report
14. Stop

question introduced, b is the distance on the carrier from the edge of the cell to the place where the error was discovered, and Text 2 is the form of the error.

At the present time the program handles 15 different types of errors. The number of such error reports can be increased as necessary.

Information from one report that had undergone syntactic analysis is subjected to statistical treatment and memorized in the information bank. The results of statistical treatment are printed out and distributed to expert analysts. When necessary the information contained within the given report is corrected as specified by the expert. The figure above shows a block diagram of the input subsystem.

The method proposed here for organizing computer input of information, ensuring its logical and syntactic analysis, and providing for error correction may enjoy extensive application in automated (computer-based) systems for solving various problems in agricultural science and practice.

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11004
CSO: 1870

AGROTECHNOLOGY

UDC 631.22.018

THE POSSIBILITIES FOR USING PIG MANURE TO GROW MICROORGANISMS

Moscow DOKLADY VSESOYUZNOY ORDENA LENINA AKADEMII SEL'SKOKHOZYAYSTVENNYKH NAUK IMENI V. I. LENINA in Russian No 3, 1979 pp 26-28

[Article by Cand Biol Sci A. V. Khotyanovich, Dr. Biol Sci Yu. M. Voznyakovskaya, Cand Biol Sci N. P. Avrova, Dr. Biol Sci A. N. Mosolov, and Cand Biol Sci Yu. Ye. Konev, All-Union Scientific Research Institute of Agricultural Microbiology, Novosibirsk Medical Institute, and All-Union Scientific Research Institute of Antibiotics and Enzyme Preparations]

[Text] Large amounts of manure are accumulating owing to organization of pig raising complexes. The main way it is utilized today is as fertilizer. This requires a great deal of work and considerable outlays on transportation, on mechanizing the means for introducing it into the soil, and so on (1).

Other ways for utilizing pig manure that consider the need for protecting the environment from polution are associated with the activities of microorganisms. Use of manure as a nutrient medium in which to grow microorganisms with the goal of obtaining microbial protein that could be fed to animals is one of the aspects of the problem of utilizing the liquid fraction of pig manure.

We know that a cubic meter of pig manure contains up to 100-140 kg dry matter, 50-70 kg organic compounds, 4.5-5 kg nitrogen, 1.5-2.5 kg phosphorus, 3-5 kg potassium, 3 kg calcium, and 1.8 kg magnesium (2).

We isolated 469 cultures of microorganisms from fresh manure from pigs, horses, cows, and sheep, from different soils, from the wastes of a meat packing plant, and from manure storehouses; some of these cultures were also borrowed from the collections of the All-Union Scientific Research Institute of Agricultural Microbiology and the All-Union Scientific Research Institute of Antibiotics and Enzyme Preparations. They included 142 bacterial cultures, 37 yeast-like cultures, 94 actinomycetes, and 196 fungi belonging to 64 genera. Nutrient medium was prepared from pig manure in the following way: Natural manure was diluted with a 1 percent

NaCl solution at a 1.3 ratio, sterilized for 1 hour at 120°C, and then kept for 2 hours at 100°C in an air current to permit deodorization. Then the solid fraction was removed with a separator.

Before using the medium we neutralized it by an addition of 20 percent NaOH solution until a pH of 7-7.2 was reached, and we sterilized it for 30 minutes at 1 gauge atmosphere. No additional nutrients were introduced into the medium.

Medium prepared in this way contained (mg/percent): Hydrolyzable carbohydrates--60, reducing sugars--15.6, total nitrogen--129.5, to include ammonium nitrogen--44.1.

Initially all microorganisms were seeded in test tubes containing medium of the indicated composition solidified with agar.

After the first passage we discovered that some of the cultures would not grow on this medium, while a number of others died in the second and third passages. In all we discarded 328 cultures, or 70 percent of the total number.

Among microorganisms that would not grow on medium made from pig manure, most had been isolated from soil, or they were fungi and bacteria from various places. Out of 142 bacterial cultures, 54 (38 percent) survived. Out of 196 cultures, only 2 were capable of growing satisfactorily in the experimental medium--*Coprinus* sp., *Myrotheaium verrucaria*.

To determine the capability microorganisms growing in the liquid nutrient medium have for accumulating biomass, we poured 100 cm^3 into 750 cm^3 flasks, sterilized the medium, and seeded with suspension obtained by washing 3-day cultures from test tubes containing shaved agar. The cultures were grown in a rocking device at 230 rpm and a temperature of 24°C for 3 days. To determine the quantity of biomass we centrifuged the fermentation liquid at 6,000 rpm, transferred the precipitate into weighing bottles, and dried them until constant weight was achieved. In all we tested 136 cultures, to include 54 bacterial, 27 yeast, and 55 actinomycete cultures. The results showed that the cultures differed in their growth activity on the experimental medium (Table 1).

Table 2 provides greater detail on accumulation of biomass by different cultures. It follows from this table that the capability for utilizing manure nutrients depends on the type of microorganism, and not on the place from which it was isolated.

Table 1. Biomass Accumulation by Different Microorganisms on Pig Manure Medium

Microorganisms	Number of Cultures Accumulating Biomass (gm/liter)			
	1--2	2--3	3--4	4--5
Bacteria	22	21	11	0
Yeast-like microorganisms	7	15	4	2
Actinomycetes	14	13	7	21

Actinomycetes developed most actively in the manure, which agrees with data published by Weiner and Rhodes (3), who grew these microorganisms in waste water from cow barns.

It follows from the data presented here that the relatively weak accumulation of biomass by all tested microorganisms growing on manure medium may be explained by the low concentration of soluble carbohydrates in the medium (not one of the tested microorganisms, except for fungi, was capable of utilizing cellulose). This is why the microorganisms were grown in the next series of experiments with the described medium supplemented by 1 percent sucrose. The results of these experiments are shown in Table 3.

Although the addition of sucrose increases biomass accumulation, the economic factor (the ratio of the amount of carbohydrates utilized to the amount of biomass accumulated) remains extremely low. As we know, this factor is usually 40-50 percent for most microorganisms grown in favorable mediums. It our experiment it varied within 4.8 and 19 percent. Consequently the reason for low biomass accumulation lies not only in a carbohydrate deficiency but also in poor utilization of carbohydrates by all microorganisms capable of existing in manure medium. Perhaps the reason for weak growth could be found in the presence of toxic substances in the manure, inhibiting productive metabolism of these microorganisms.

Thus we established that out of all investigated taxonomic groups of microorganisms (yeasts, fungi, bacteria, actinomycetes) capable of growing in manure, actinomycetes grow and accumulate biomass most actively. At the same time biomass accumulation does not attain the level typical of these microorganisms when grown in favorable mediums, which can probably be explained by presence of toxic substances in mediums prepared from pig manure. Total biomass accumulation in microorganism cultures remains extremely low. All of this information indicates that use of manure as a substrate for microbial synthesis of nutrient protein is unpromising.

Table 2. Cultures of Microorganisms Growing Most Actively on Pig Manure Medium

№ культуры (1)	Систематическое положение (2)	Коллекционный номер штамма (3)	Откуда выделены или получены (4)	Вес сухой биомассы (г/л) (5)
68	Microbacterium	Оригинальный (8)	Коровий навоз (9)	3,91
83	»	»	Конский навоз (10)	2,87
102	Micrococcus	» (9)	Коровий навоз	3,09
130	Sarcina lutea	18/978		3,87
131	то же (6)	17/220	Музей ВНИИ СХМ (11)	3,95
132	»	16/225		2,77
134	Sarcina aurantiaca	11/981		3,04
20	Bacillus	Оригинальный (8)	Чернозем (12)	2,73
113	»	»	Стоки мясокомбината (13)	3,67
114	»	»	То же (6)	3,90
115	»	»	» »	3,78
117	»	»	Жировой завод (14)	3,69
77	Дрожжи (7)	Оригинальный (8)	Свиной навоз (15)	4,11
79	»	То же (6)	То же (6)	3,15
175	»	68	Жировой завод (14)	3,78
26	Actinomyces	Оригинальный (8)	Чернозем (12)	4,10
119	Act. coelicolor	13/937	Музей ВНИИ СХМ (11)	4,29
123	Actinomyces	8/184	То же (6)	4,40
124	Act. erythreus	7/182	» »	4,40
126	Act. diastochromogenes	4/175	» »	4,95
127	Act. citreus	3/173	» »	3,57
164	Act. griseus	15	» »	4,00
165	Actinomyces	0191	» »	4,36
210	»	0027	Ин-т антибиотиков (16)	5,15
213	»	0690	То же (6)	5,20
221	Act. Griseus	0540	» »	4,36
226	Actinomyces	0094	» »	4,62
231	»	0166	» »	5,70
234	»	0691	» »	4,66
182	Дрожжи (7)	3	Жировой завод (14)	3,65

Key:

1. Culture number
2. Taxonomy
3. Strain collection number
4. Place from which isolated or obtained
5. Dry biomass weight (gm/liter)
6. As above
7. Yeast
8. Original
9. Cow manure
10. Horse manure
11. Museum of the All-Union Scientific Research Institute of Agricultural Microbiology
12. Chernozem
13. Meat packing plant wastes
14. Tallow plant
15. Pig manure
16. Institute of Antibiotics

Table 3. Effect of Readily Available Carbon Source on Growth of Some Microorganisms on Pig Manure Medium

(1) Микроорганизмы	(2) № культуры и место выделения	(3) Биомасса на исходной среде, (г/л)	(4) Биомасса на среде + + 1% сахарозы (г/л)	Экономический коэффициент использования сахарозы (%) (5)
Бактерии (6)	Microbacter, 68, из коровяка (9)	1,38	2,24	8,6
	Bacillus, 114, из стока мясокомбината (10)	2,10	2,58	4,8
Дрожжеподобные (7)	77, из свиного навоза (11)	2,37	3,42	10,5
	5, из стоков мясокомбината (10)	4,34	5,61	12,7
Актиномицеты (8)	231, из Ин-та антибиотиков (12)	2,80	4,70	19,0
	39, из серозема (13)	2,80	4,38	15,8

Key:

1. Microorganisms
2. Culture number and place of isolation
3. Biomass on initial medium (gm/liter)
4. Biomass from medium + 1% sucrose (gm/liter)
5. Economic sucrose use factor (%)
6. Bacteria
7. Yeast-like microorganisms
8. Actinomycetes
9. From liquid cow manure
10. From meat packing plant wastes
11. From pig manure
12. From Institute of Antibiotics
13. From serozem

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11004
CSO: 1870

AGROTECHNOLOGY

UDC 546.47+546.48:543.53

DETERMINATION OF ZINC IN THE PRESENCE OF CADMIUM BY ISOTOPIC DILUTION COUPLED WITH SUBSTOICHIOMETRIC ISOLATION

Moscow DOKLADY VSESOYUZNOY ORDENA LENINA AKADEMII SEL'SKOKHOZYAYSTVENNYKH NAUK IMENI V. I. LENINA in Russian No 3, 1979 pp 28-30

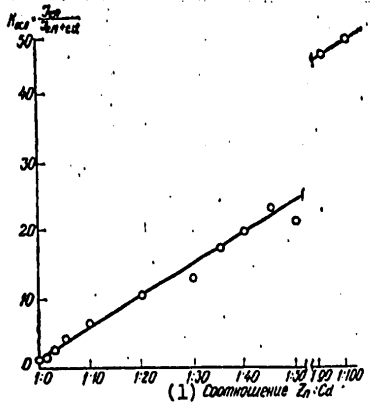
[Article by Cand Chem Sci I. Ye. Zimakov, All-Union Scientific Research Institute of Veterinary Medicine]

[Text] We know that cadmium (in compound form) is a highly toxic element. Its chemical analog--zinc--is a vitally necessary microelement to plants and animals. But high concentrations of zinc in soil, and correspondingly in animal feed and food products cause serious illness in animals and man. Zinc contamination may be especially high in regions of developed metallurgical and chemical industry. In this case the zinc is usually found together with cadmium. In many cases the need arises for determining these elements separately.

Determination of zinc in the presence of cadmium is a complex analytical task in view of the similarity of their chemical properties, for example the extraction constants of their compounds (5). To perform an analysis we would usually have to first separate the elements (1), which results in a certain loss of the elements, causing the results to become distorted. Attempts have been made at determining relatively large quantities of zinc in cadmium preparations by a radiometric method making use of calibration curves (8,9).

This paper examines the possibility for determining microquantities of zinc in the presence of cadmium without their preliminary separation by capitalizing upon the linear dependence, which we discovered earlier (7), of attenuation of the scintillation intensity of substoichiometrically isolated samples on the Cs:K ratio for the solution under analysis. In this regard it would be interesting to establish the law governing this dependence in relation to other pairs of elements with similar chemical properties, using zinc and cadmium as the example, and, on the basis of the obtained results, to develop the method for determining zinc in the presence of cadmium.

Experiments were performed on extractional-substoichiometric isolation from solutions containing just zinc alone and from solutions containing the same quantities of zinc and different quantities of cadmium. The conditions of substoichiometric isolation of zinc and cadmium dithizonate (2,3) were as follows: The pH of the solution under analysis was set at 8±0.5, the samples were shaken hard in a separatory funnel for 5 minutes after addition of a substoichiometric quantity of dithizone in chloroform (usually 75 percent of the stoichiometric quantity of zinc) (4).



Graphical dependence of the attentuation factor of the scintillation intensity of substoichiometrically isolated samples on the Zn:Cd ratio for the extracted solution: $K_{осл}$ --attenuation factor; J_{Zn}--scintillation intensity (pulses/min) of the organic phase upon extraction from the solution containing zinc alone; J_{Zn+Cd}--scintillation intensity (pulses/min) of the organic phase upon extraction from solutions containing the same quantity of zinc and added cadmium

Key:
1. Zn:Cd ratio

As we can see from the figure we obtained a line which intersects the ordinate at a point corresponding to the ratio Zn:Cd=1:0. We established that the attenuation factor for the scintillation intensity of substoichiometrically isolated samples has a linear dependence on the ratio of zinc to cadmium in the extracted solution. Basing ourselves on the obtained data we can suggest a method for determining microquantities of zinc in samples having a known cadmium concentration, the essence of which is as follows. The sample to be analyzed is dissolved, and two identical aliquots are taken, containing x µg zinc and mx µg cadmium. In this case mx is known while the ratio $x:mx=1:m$ is unknown. A quantity of cadmium that is a

Determination of Zinc by Isotopic Dilution in the Presence of Cadmium

(1) Содержание (мкг)			(5)				(6)	(7)	(8)	(9)	(10)	(11)
цинка в анализируемом растворе (2)	кадмия в анализируемом растворе (3)	кадмия во второй аликвоте части (4)	Соотношение Zn:Cd в анализируемом растворе	n	K_m	K_{mn}	Найденное соотношение Zn:Cd в первой аликвоте (по графику)	Рассчитанное содержание Zn в первой аликвоте (мкг)	Найденное соотношение Zn:Cd во второй аликвоте (по графику)	Рассчитанное содержание Zn во второй аликвоте (по графику)	Среднее значение (мкг)	Ошибка определения (отн. %)
5,00	5,00	10,0	1:1	2	1,48	1,96	1:1,1	4,55	1:1,9	5,26	4,91	1,8
5,00	5,00	50,0	1:1	10	1,45	5,48	1:0,95	5,26	1:9,9	5,05	5,16	3,2
5,00	5,00	125,0	1:1	25	1,50	13,46	1:1	5,00	1:26,9	4,65	4,83	3,4
0,50	5,00	10,0	1:10	2	5,78	10,57	1:10	0,50	1:20,3	0,49	0,49	2,0
0,50	5,00	25,0	1:10	5	5,77	24,89	1:10	0,50	1:50,3	0,49	0,49	0,0
0,50	5,00	50,0	1:10	10	4,50	31,50	1:8	0,62	1:105	0,47	0,54	8,0
0,050	2,50	5,0	1:50	2	19,6	29,90	1:40	0,061	1:102	0,049	0,055	10,0

Key:

1. Concentration (μg)
2. Zinc in analyzed solution
3. Cadmium in analyzed solution
4. Cadmium in second aliquot
5. Zn:Cd ratio in analyzed solution
6. Found Zn:Cd ratio for first aliquot (from graph)
7. Computed Zn concentration in first aliquot (μg)
8. Found Zn:Cd ratio in second aliquot (from graph)
9. Computed Zn concentration in second aliquot (from graph)
10. Mean (μg)
11. Determination error (relative percent)

multiple of mx is added to the second aliquot. In this case the total cadmium concentration in it would become equal to nmx, and correspondingly the Zn:Cd ratio would equal $1:n \cdot m$, where n is a number indicating how many times more cadmium there is in the second aliquot than in the first. Next an equal quantity of ^{65}Zn isotope is added without the carrier to both aliquots of the solution under analysis, the pH is adjusted to 8±0.5, and zinc (combined with cadmium) is subjected to substoichiometric isolation by means of extraction by chloroform containing identical substoichiometric quantities of dithizone. Then we measure the scintillation intensity of the organic phases, J_1 and J_2, and we compute the attenuation factor for the first and second aliquots (K_m and K_{nm}) using the formulas:

$$K_m = 1 + \frac{J_1 - J_2}{nJ_2 - J_1}, \qquad (1)$$

$$K_{nm} = 1 + \frac{n(J_1 - J_2)}{nJ_1 - J_2}, \qquad (2)$$

derived from an examination of similar triangles constructed in arbitrary coordinates of the dependence of the attenuation factor of the scintillation intensity of substoichiometrically isolated samples on the Zn:Cd ratio for the extracted solutions (7).

Having computed K_m and K_{mm} with formulas (1) and (2), we find the Zn:Cd ratio for both aliquots using the graph plotted on the basis of experimental results (see figure). Knowing the quantity of cadmium in the samples, we can compute their zinc concentrations.

When the zinc concentration in the aliquots is less than 0.01 μg, in order to keep from overstepping the bounds of the determination method's sensitivity (6)--that is, in order to keep the concentration of zinc in the aliquot subjected to extraction greater than or equal to m_{min}--an equal quantity of zinc is added to both aliquots in the form of a radioactive standard solution (0.01 μg in our case), which in turn increases the method's sensitivity.

The table shows the results of determining zinc in the presence of cadmium using model solutions.

This method may also be used to analyze solutions containing just zinc alone, without cadmium. For this purpose an identical quantity of ^{65}Zn is added without the carrier to two equal aliquots of the solution under analysis, each aliquot containing x μg Zn, and a standard cadmium solution is added to one of them in a quantity resulting in a Zn:Cd ratio approximately within the range from 1:1 to 1:100. The subsequent steps of the analysis and computations are as described above.

Thus capitalizing on the linear dependence of attenuation of the scintillation intensity of substoichiometrically isolated samples on the ratio of elements having similar chemical properties in the solution under analysis (where the concentration of one of the elements is known), we have developed a method for determining zinc in the presence of cadmium.

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11004
CSO: 1870

AGROTECHNOLOGY

UDC 633.18:631.52

A STUDY OF PLANTS REGENERATED IN RICE TISSUE CULTURE

Moscow DOKLADY VSESOYUZNOY ORDENA LENINA AKADEMII SEL'SKOKHOZYAYSTVENNYKH NAUK IMENI V. I. LENINA in Russian No 3, 1979 pp 10-12

[Article by Cand Ag Sci L. A. Kucherenko and G. G. Mamayeva, All-Union Scientific Research Institute of Rice]

[Text] The method of growing isolated tissues does not just serve the needs of theoretical research in biology alone; it can also be used for plant selection purposes (1).

The level of spontaneous mutagenesis rises dramatically in cultured cells. The methods for obtaining callus tissue and regenerating plants from it have been developed for most agricultural plants. By regenerating plants from cells altered in the culturing process, we can hope to obtain genetically altered forms of plants that would serve as the raw material for selection (5).

An increase in the level of spontaneous mutagenesis *in vitro* has been noted in a number of works. Sometimes it attains very high values. Thus the cell mutation level was 6-7 percent in the first passages of *Haplopappus officinalis*, and it was up to 70 percent in a 4-year culture of tobacco tissue (4).

Works describing regenerants arising in tissue culture are still few in number, and the research has been performed only in relation to a small number of plant species (2,6-10).

Unfortunately not one of these works deals with the behavior of regenerants in subsequent generations. We decided to study two generations of plants regenerated from rice callus tissue.

We used four rice varieties in our work--VNIIR 3970, VNIIR 3995, VNIIR 3973, and VNIIR 3942. We obtained callus tissue in 1977 on (Murasigi-Skug) agar nutrient medium containing 2,4D using the embryos of mature seeds from these varities. After substituting 2,4D by indolylacetic acid and kinetin the callus

regenerated sprouts which, following transplanting into soil-filled containers, produced adult plants. This generation was designated P_1. Regeneration was performed after four passages of the callus for variety VNIIR 3995, while for the rest of the varieties the plants were regenerated from the first passage.

Seeds collected from every separate P_1 plant were sown in spring 1978 in an outdoor nursery. For comparison purposes we planted seeds from the initial varieties, obtained by the usual means. Phenological observations were made during the course of vegetation. After the plants matured, they were all subjected to meticulous inspection, and visual differences from the standards were described, after which we performed biometric analysis on the basis of the following characteristics--plant height, bushiness, panicle productivity, and the frequency of empty grains.

In all in 1977 we obtained 180 P_1 sprouts. Some of them were weak, especially those from test tubes in which 5-10 and more of them came up right away, and after being transplanted into soil they died; most of the sprouts, however, did produce adult plants.

It was difficult to compare P_1 plants among each other and with the initial varieties because the time of regeneration of plants from callus tissue is long: Plant sprouting proceeds for 2.5 months from the same passage. Therefore the regenerants were planted at different calendar times, out in the open or in protected soil depending on the time of the year, which could not but have a reflection on a number of characteristics (plant height, productivity and sterility of panicles, and so on).

Moreover sprouts that had regenerated in quantities of not more than one or two from each callus had a clear advantage over others appearing in large groups (sometimes 10 and more from a single test tube) due to better nutrition in the first phases of development. In addition the latter were so close together that they were injured when an attempt was made to separate them, thus weakening the plants even more because of the limited space available in controlled climate chambers; the plants were grown in small soil volumes, which also had an effect on their growth and development.

Because conditions in the initial phases of development are clearly incomparable, in our opinion it would be generally incorrect to compare the first generation of plants obtained from a callus and plants grown from seed in relation to vegetation time.

We also had to reckon with the fact that growth substances in the nutrient media (kinetin for example) may have a modifying action on plants; in particular they may cause sucker development and excessive bushiness, which disappear in subsequent generations (2).

P_1 plants differed among each other in relation to a number of characteristics, mainly the productivity and sterility of the panicle. In some plants

the main panicle did not produce grain at all, and seeds could be obtained only from side shoots or from aftergrowth. Only sporadic P_1 plants were albinos in this experiment, while most sprouts regenerating from callus material obtained from anthers were albinos (3). A number of plants exhibited profuse tillering, starting while still in test tubes. Branching of the stem (a generally rare phenomenon in rice) was a rather usual phenomenon among P_1 regenerants, and corrugated leaves were observed among some plants.

For the reasons stated above the features noted for the P_1 plants could not be used as the grounds for making conclusions concerning presence of genetically altered forms among the regenerants. Such conclusions could be made only after studying their progeny.

The seeds were obtained from 113 P_1 plants, at a quantity of from 1 to 477 per plant. These seeds were planted in a field nursery. Some of the plots that had been damaged by flooding were discarded. The result of studying the remaining 98 lines of P_2 regenerants are shown in the table and the figure.

Analysis of the P_2 generation showed that about 36 percent of the lines have morphophysiological differences from the initial varieties, in which case some of the lines are concurrently altered in relation to two and, more rarely, three characteristics. It should also be considered that we have described only the obvious phenotypic deviations. The possibility is not excluded that the outwardly, phenotypically unaltered lines may also possess differences, for example in relation to grain quality or reactions to disease or various environmental factors, which may be revealed by appropriate analysis.

The high percentage of altered lines among the regenerants attracts attention. For comparison purposes we present data obtained for induced mutagenesis achieved by the conventional method: Seeds from variety VNIIR 3995 were treated with γ-rays in 1977. Low germination of irradiated seeds and the high sterility of M_1 plants meant that the dose was close to lethal. In 1978 we planted 500 M_2 lines. Mutants (mainly short forms) were found in 24 plots, which corresponded to 4.8 percent of the total number of lines.

Presence of lines with altered phenotype in the second generation after transplanting of the regenerants indicates that the changes are genetic in nature, and that they are obviously the result of spontaneous mutations. The features noted in the P_1 generation, meanwhile--dramatically greater bushiness, branching of the stem, and corrugated leaves--did not appear in the P_2 generation, which confirms the notion that these were nongenetic modifications.

The material obtained in this experiment was not subjected to cytogenetic analysis. But judging from the phenotype, all lines were diploid: Not one line with high sterility or other typical characteristics indicating possible alteration of ploidy was found in the P_2 generation.

Results From Studying Second-Generation Regenerants Obtained From Rice Callus Tissue

(1) Сорт	(2) Пассаж, из которого получены регенеранты	Количество линий P_1 (3)			Основные отличия от исходного сорта (7)		
		(4) всего	в т. ч. с измененным фенотипом (5)		(8) признак	(9) количество линий с различными типами изменений	из них с расщеплением по данному признаку (10)
			шт. (6)	%			
ВНИИР 3995 (11)	I	13	2	15,4	Более позднее созревание (13)	1	1
					Низкорослость* (14)	2	1
					Наличие на метелке абортивных веточек (15)	1	1
	II	6	1	16,7	Низкорослость (14)	1	—
	IV	7	3	42,9	Низкорослость (14)	2	1
					Более позднее созревание (13)	2	1
					Наличие на метелках абортивных веточек (15)	1	1
ВНИИР 3970	I	48	18	37,5	Низкорослость (14)	12	—
					Наличие на метелках абортивных веточек (15)	1	1
					Более продуктивная метелка (16)	3	2
					Повышенная устойчивость к слою воды** (17)	3	—
					Остистость (18)	1	1
ВНИИР 3973	I	17	11	64,6	Низкорослость (14)	2	—
					Измененная форма метелки (19)	4	3
					Более раннее созревание	8	4
ВНИИР 3942	I	7	1	14,3	Уменьшение степени опушенности зерна (20)	1	1
Итого: (12)	—	98	36	36,7	—	—	—

*Shortness was accompanied in all cases by lower panicle productivity, but the degree of this decline varied significantly among different lines.

**Hypothesized from the behavior of plants in the field when deeply flooded; requires checking in a special experiment.

Key:

1. Variety
2. Passage from which regenerants were obtained
3. Number of P_2 lines
4. Total
5. To include with altered phenotype
6. Number
7. Basic differences from initial variety
8. Characteristic
9. Number of lines with different types of changes
10. Of these, those exhibiting variations in relation to this characteristic
11. VNIIR
12. Total
13. Later maturation
14. Shortness*
15. Presence of abortive sprigs in the panicle
16. More-productive panicle
17. High resistance to standing water**
18. Presence of awns
19. Altered panicle shape
20. Reduced grain silkiness

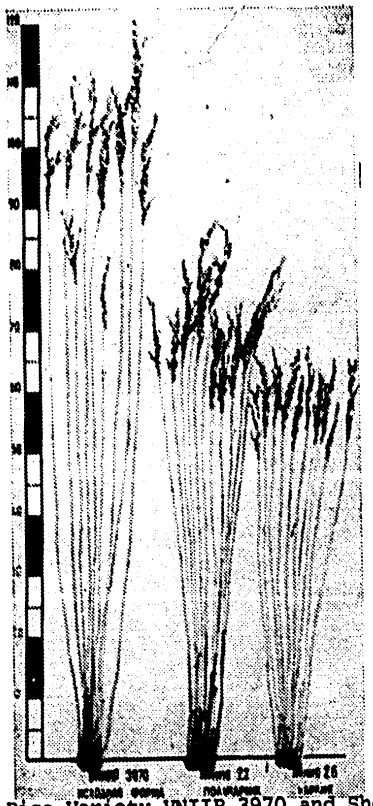

Figure 1. Rice Variety VNIIR 3970 and Short Forms Obtained From It by *in vitro* Culture

Thus by culturing somatic rice tissue and subsequently regenerating plants from it we can obtain genetically altered forms that may serve as an additional source of raw material for selection purposes.

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CSO: 1870

AGROTECHNOLOGY

UDC 633.358:576.342

INTERACTION OF ALUMINUM AND PHOSPHORUS ON THE ROOT SURFACE AND IN CELL WALLS

Moscow DOKLADY VSESOYUZNOY ORDENA LENINA AKADEMII SEL'SKOKHOZYAYSTVENNYKH NAUK IMENI V. I. LENINA in Russian No 3, 1979 pp 6-8

[Article by VASKhNIL Corresponding Member E. L. Klimashevskiy, Cand Biol Sci Yu. A. Markova, and I. S. Lebedeva, Siberian Institute of Plant Physiology and Biochemistry of the Siberian Branch of the USSR Academy of Sciences, and the Siberian Institute of Chemicalization of Agriculture of the Siberian Branch of VASKhNIL]

[Text] A knowledge of the nature of phosphorus absorption in light of ionic toxicity and of its fixation by aluminum in the first stage of uptake of these substances by plants might bring us closer to a better understanding of the causes behind aluminum toxicity (1,2,6).

Studying the roots of two varieties of peas (Tulun green--sensitive to aluminum, and Uspekh--resistant to it), we determined the quantity of aluminum absorbed by the root hair zone (RHZ) and by cell walls (CW) within this zone, as well as the quantity of aluminum taken up by root epidermis and by tissues with the epidermis removed in 1 hour from solution containing $2.5 \cdot 10^{-4}$M Al^{3+} .

After the plants and isolated CW were kept in a solution containing $5 \cdot 10^{-3}$M phosphorus for 1 hour, we determined the quantity of inorganic phosphorus in the root zone and in CW. We selected 40 sprouts for our experiments, from which we first removed the cotyledons; these sprouts were placed in 100 ml $AlCl_3$ for 1 hour. Then the roots were dried on filter paper and placed in distilled water (100 ml) for 30 seconds to remove the surface film of ions. After this the plants were placed in KH_2PO_4 solution for 1 hour; then they were dried and the surface film was removed. RHZ were taken for analysis from roots processed in this way. In the case where we used epidermis and root tissue without epidermis from the RHZ, we removed the upper cell layers with forceps (3). The experiments with isolated CW were conducted as were the root experiments. The CW material was weighed out in 0.25 gm samples.

In order that we could create conditions in the experiments with isolated CW that were identical to those of the root experiments, we set the volume of incubation solution at 90 ml. We established that 1 gm (wet weight) of RHZ from both varieties contained 0.0793 gm (wet weight) CW, while RHZ material from 40 roots weighed 3.5 gm (wet weight).

All solutions used in the experiments were adjusted to pH 4, as was true with the distilled water used on control samples.

The ionic surface film was removed with distilled water having an initial pH of 5.7. The experiments were performed at 22-23°C. Experimental replication was threefold. The method for isolating CW and determining aluminum was described earlier (5). Inorganic phosphorus was determined by Lowry's method as modified by V. P. Skulachev. The results of the experiments were statistically treated by V. L. Voznesenskiy's method.

RHZ material from the two varieties accumulated different quantities of aluminum following a 1 hour exposure of the plants to the solution containing Al^{3+} (Table 1). Presence of Al ions in the solution significantly increased the quantity of inorganic phosphorus in RHZ cells. The variety sensitive to Al^{3+} toxicity absorbed less phosphorus than in the control variant, but it accumulated more phosphorus in the presence of aluminum than did plants of the tolerant genotype. RHZ material from the sensitive variety increased its capability for fixing phosphorus by 3 times after being processed with Al ions, while material from the resistant variety increased its capability by 1.3 times (Figure 1).

Table 1. Quantity of Aluminum Absorbed by RHZ Material (mg/gm Dry Weight per Hour)

Variety	Experimental Variant	Al Concentration	Al Absorbed
Tulun green	without Al with Al	0.28±0.00 0.96±0.03	0.68
Uspekh	without Al with Al	0.14±0.00 0.95±0.004	0.81

Phosphate ions did not settle on the surface of isolated CW material in the variant without aluminum. Following adsorption of Al ions, the CW material did fix phosphorus ions (Table 2). The affinity of aluminum to pectin substances is known (4). Apparently when the H+ ions of pectin carboxyl groups are substituted by Al ions, the latter bind with CW polysaccharides through one of the available protons. It could be hypothesized that the remaining valent bonds may be occupied by PO_4 anions. This may explain concentration of these anions in response to processing of the CW material with Al^{3+} (6).

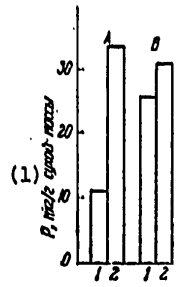

Figure 1. Effect of Al on P Absorption by the Root Hair Zone of Peas: *A*--Tulun green; *B*--Uspekh; *1*--variant without Al; *2*--with Al

Key: 1. P, μg/gm dry weight

Table 2. Effect of Aluminum on Phosphorus Absorption by RHZ Material (μg/gm Wall Dry Weight)

(1) Сорт	(2) Вариант опыта	(3) Содержание P	Поглощено (4) P
Тулунский зеленый (5)	H_2O+H_2O	364,8±1,2	11,3
	$H_2O+KH_2PO_4$	376,1±1,4	
	$AlCl_3+H_2O$	368,6±8,3	33,9
	$AlCl_3+KH_2PO_4$	402,5±18,0	
Успех (6)	H_2O+H_2O	305,3±3,6	25,6
	$H_2O+KH_2PO_4$	330,9±5,3	
	$AlCl_3+H_2O$	113,0±13,8	33,4
	$AlCl_3+KH_2PO_4$	346,3±11,4	

Key:
1. Variety
2. Experimental variant
3. P concentration
4. P absorbed
5. Tulun green
6. Uspekh

Eighty-seven percent of the absorbed phosphorus is deposited in walls of the RHZ from the sensitive form of peas, while for the resistant form the figure is 94 percent. Thus phosphorus is precipitated by aluminum outside the protoplast. Apparently increasing accumulation of PO_4 ions occurs in pea roots due to adsorptive binding of Al^{3+} and P in cell walls. It is entirely possible that a certain proportion of the Al ions remain in unbound state in the double electric layer, and may form the insoluble precipitate $AlPO_4$.

The quantity of aluminum absorbed by roots and its distribution in the epidermis and in root tissues without epidermis are shown in Figure 2. As we can see, the tolerant genotype concentrates 66 percent of the aluminum in its epidermis, while the sensitive variety concentrates 82 percent.

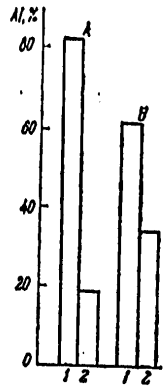

Figure 2. Quantity of Al^{3+} in Epidermis (*1*) and in the Root Hair Zone (*2*) of Peas (Percent of Total Entering the Zone): *A*--Tulun green; *B*--Uspekh

It is probable that the bulk of fixed phosphates is also contained within the epidermis. We discovered by the histochemical method suggested by Ikeda et al. (7) that roots of the wheat barley varieties that are more sensitive to acidity are stained more intensively by aluminum. Larry et al. (8) describe Al-PO_4 interaction and believe that precipitation occurs not as a continuous layer but rather in the form of point globules. Interaction occurs between aluminum and phosphorus, in their opinion, outside the cell membrane, but in the area of the root cap it is observed inside the cells. Presence of aluminum within cells of the root cap was also noted in our experiments (2).

Thus accumulation of PO_4 ions occurs due to adsorptive binding of aluminum and phosphorus in the CW, and a part of these ions are in unbound state in the double electric layer, where they form the insoluble precipitate $AlPO_4$. The greater part of the aluminum and phosphorus entering the plant within the course of an hour is retained on the root surface. Fixation of phosphorus in the CW and on the root surface during the initial stage of absorption leads subsequently to significant impairment of the absorption of phosphorus and its transport into the above-ground part of the plant, which in turn causes impairment of its metabolism in plants (2).

The hourly increase, caused by aluminum, in phosphorus uptake by the roots of the genotype sensitive to aluminum toxicity is more significant than that experienced by the resistant variety. The strength with which aluminum is fixed in the CW is rather high, it being much greater in the sensitive variety than in the resistant one (3); this is apparently what promotes firmer fixation of phosphorus in sensitive plants. Presence of aluminum on the root surface goes a long way in controlling admission of phosphorus into free spaces within the root, and limitation of phosphorus uptake means a decrease

in the transport of phosphorus into above-ground organs. Considering that the investigated pea varieties accumulate different quantities of inorganic phosphorus in the presence of aluminum and the transport of phosphorus from roots into photosynthesizing organs is retarded (2), the intensity of phosphorus metabolism and the rate of energy reactions in photosynthetic organs vary in different directions under these conditions. Consequently the resistance of individual varieties to the effect of high concentrations of free aluminum ions is closely associated with the capability the plants have for adsorbing and utilizing phosphorus in the presence of excess Al^{3+}.

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CSO: 1870

AGROTECHNOLOGY

UDC 576.851.252:546.215

STUDY OF THE ULTRASTRUCTURE AND RESPIRATORY ACTIVITY OF STAPHYLOCOCCUS IN RESPONSE TO THE ACTION OF HYDROGEN PEROXIDE

Moscow DOKLADY VSESOYUZNOY ORDENA LENINA AKADEMII SEL'SKOKHOZYAYSTVENNYKH NAUK IMENI V. I. LENINA in Russian No 3, 1979 pp 42-45

[Article by K. Sh. Dosanov, All-Union Scientific Research Institute of Veterinary Medicine]

[Text] Being a strong oxidant having high bactericidal, sporicidal, and viricidal action, hydrogen peroxide is broadly employed in the disinfection of various objects in medicine, veterinary medicine, and food industry (6,7).

The advantages of hydrogen peroxide over other disinfecting agents include the harmlessness of its breakdown products, low toxicity, and high antimicrobial activity. But the mechanism of action of hydrogen peroxide on microorganisms has not been revealed sufficiently as yet.

The objective of the present investigation was to study the ultrastructure and respiratory activity of *Staphylococcus* in response to the action of hydrogen peroxide with the goal of unraveling some aspects of this preparation's mechanism on bacteria.

Eighteen-hour *Staph. aureus*, strain Nikitin grown on beef-extract agar was used as the test object. A bactericidal suspension containing $2 \cdot 10^9$ cells/ml was prepared from the culture.

Staphylococcus survival following exposure to hydrogen peroxide was determined by dilution followed by agar seeding and the counting of the resulting colonies. The obtained data were used to derive survivability curves, which were placed at the basis of our study of structural and functional changes occurring in the bacteria (3).

Preparations were prepared for electron microscopy in accordance with the commonly accepted techniques (10) and embedded in an (epon-araldit) mixture. Ultrathin sections obtained with an LKV-4801 A ultratome and stained with uranyl acetate and lead citrate (9) were viewed with a Hitachi-12 electron microscope.

Bacterial respiratory activity was determined manometrically with Warburg apparatus by the method suggested in (2) as modified by us. This is why before measuring the respiration level of the bacteria with the Warburg apparatus we carefully washed hydrogen peroxide from the cells with 0.02 M phosphate buffer and centrifugation. Then the bacteria were transferred to Warburg vessels, and their respiratory activity was determined (2). Cell oxygen consumption was measured three times at 30 minute intervals.

Analysis of the dynamics of *Staphylococcus* death (*in vitro*) showed that a 0.6 percent hydrogen peroxide solution caused the death of 10 percent of the cells in 2-3 minutes (T_{10}), 50 percent after 5-7 minutes (T_{50}), and 90 percent after 50-55 minutes (T_{90}) (Figure 1*a*).

Electron microscopic analysis of the ultrastructure of *Staphylococcus* before exposure to hydrogen peroxide (control) showed that the cells (Figure 2*a*) had a smooth, homogeneous cell wall with a thickened inner layer, and a single-layered cytoplasmic membrane. The cytoplasm was densely packed with granular components--ribosomes, and an osmiophobic nucleoid in the central part of the cell.

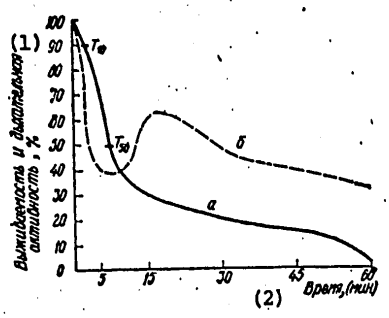

Figure 1. Mortality Dynamics and Change in Respiratory Activity of *Staphylococcus* in Response to 0.6 Percent Hydrogen Peroxide Solution: *a*--Survival; *b*--Respiratory Activity

Key:
1. Survival and respiratory activity, percent
2. Time, minutes

Immediately after exposure to hydrogen peroxide (T_{10}) a certain increase of the volume of the cells (without visible disturbance of the cell wall's integrity), cytoplasmic membrane, cytoplasm, and nucleoid were noted in the ultrastructure of *Staph. aureus* (Figure 2*b*). Longer exposure to hydrogen

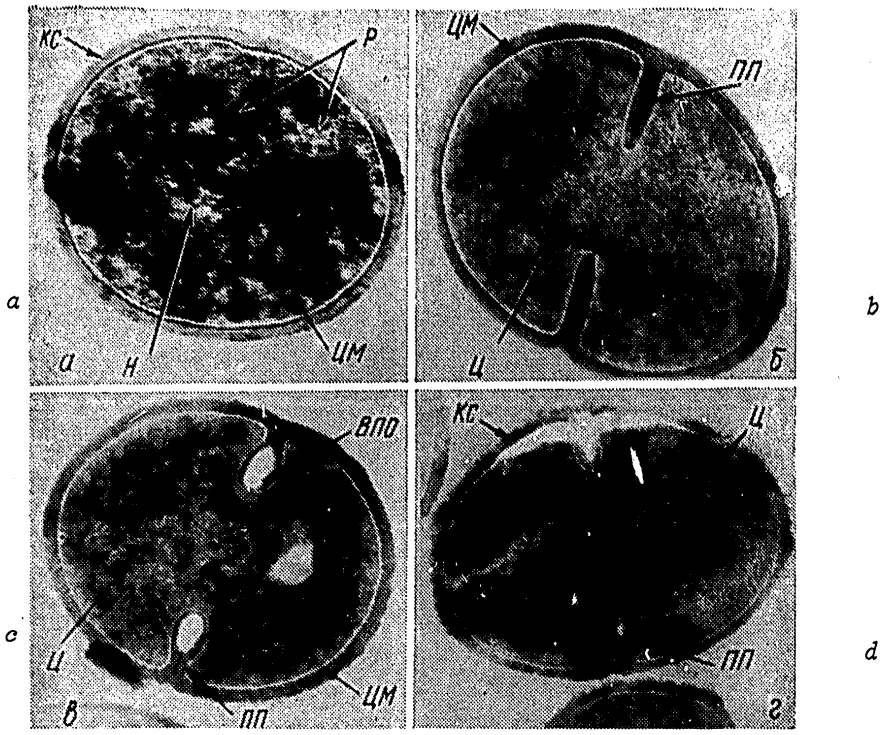

Figure 2. Action of Hydrogen Peroxide on *Staphylococcus*: *a*--Ultrastructure of *Staph. aureus* (control), ×65,000; *b*--initial action (T_{10}); *c*--sublethal (T_{50}); *d*--lethal (T_{90}); КС--cell wall; ЦМ--cytoplasmic membrane; ПП--transverse septum; Ц--cytoplasm; Р--ribosomes; ВПО--vacuole-like formations; Н--nucleoid

peroxide (T_{50}) caused fragmentation and insignificant swelling of the cell wall of *Staph. aureus* cells. Some of the segments of the cytoplasmic membrane remained invisible. Vacuole-like formations were discovered in the cytoplasm and in the transverse septums of individual cells (Figure 2*c*). When *Staph. aureus* was subjected to a lethal disinfectant dose (T_{90}) we distinctly observed local lesions of the cell wall and the cytoplasmic membrane. Electron-opaque zones were visible in the cytoplasm; the polyribosomes and ribosomes were not clearly differentiated (Figure 2*d*).

Manometric analysis of respiratory activity showed that control (untreated) *Staphylococcus* cells consumed 60 µl oxygen.

When *Staphylococcus* was exposed to 0.6 percent hydrogen peroxide solution for 5 minutes the respiratory activity of the cells decreased by 40 percent of the initial level, while exposures of 15 and 60 minutes caused corresponding decreases of 63 and 31 percent (Figure 1*b*).

When hydrogen peroxide breaks down it releases active atomic oxygen and highly reactive radicals (7,8).

The enlargement of *Staph. aureus* cells and the dramatic inhibition of respiratory activity upon initial exposure (T_{10}) to hydrogen peroxide are probably associated with the cell's excessive uptake of hydrogen ions formed by dissociation of hydrogen peroxide, resulting in hydration of the bacteria. Our hypothesis agrees with the data of other researchers (5) who observed hydration of microbial cells following exposure to peracetic acid.

Apparently the enlargement of cell volume following exposure to hydrogen peroxide occurs due to disturbance of the permeability barrier of the cell wall and cytoplasmic membrane of *Staphylococcus*. Consequently it becomes easier for the products of hydrogen peroxide breakdown to enter the cell.

When oxygen and free hydroxyl radicals enter the cell in sufficient quantities, excessive peroxidic oxidation of lipids occurs (1). We know that the bulk of the lipids of Gram-positive bacteria, including *Staphylococcus*, is concentrated in the cytoplasmic membrane. Moreover it contains structural proteins and various enzymes, particularly redox enzymes. Hence we can assume that the principal place of peroxidic oxidation of lipids in the microbial cell is the cytoplasmic membrane, which performs many vitally important functions, including respiratory functions.

Active oxygen and free hydroxyl radicals formed by breakdown of hydrogen peroxide apparently interact mainly with membrane proteins and lipids, which is manifested externally by structural changes in the membranes and has an inhibitory effect on respiration by cells subjected to the action of hydrogen peroxide.

Our data confirm the point of view suggested by Vladimirov and Archakov (1), who believe that excessive oxidation of lipids leads to dramatic disturbance of the physicochemical structure of membranes, going as far as their complete rupture.

Some researchers (1,5) believe that atomic oxygen liberated in contact with the microbial cell interacts with the most sensitive protein groups, for example the SH groups of cysteine, and subsequently they destroy other bonds. It is interesting to note that inhibition of respiratory activity occurred in stages in response to hydrogen peroxide exposure. It was noted to decline dramatically in the first minutes after exposure to the preparation, after which it increased somewhat and then declined once again.

In my opinion the short-term rise in respiratory activity following sharp inhibition is probably associated with partial release of enzymes from the cell as a result of the greater permeability of the cell wall and the cytoplasmic membrane, leading to an improvement of substrate-enzyme interaction. Further inhibition of respiration is apparently associated with destruction of the cytoplasmic membrane and with the inhibitory action the products of peroxidic oxidation (alcohols, ketones, aldehydes, and so on) have on enzymes.

Our hypotheses agree with the opinions of other researchers (4) who have noted stimulation of the activity of respiratory enzymes following exposure to low concentrations of disinfecting agents.

Thus our research showed that 0.6 percent is the lethal concentration of hydrogen peroxide for *Staphylococcus (in vitro)*. In this case the cell mortality dynamics correlate completely with inhibition of respiratory activity only in the first 5-7 minutes (T_{10}, T_{50}).

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